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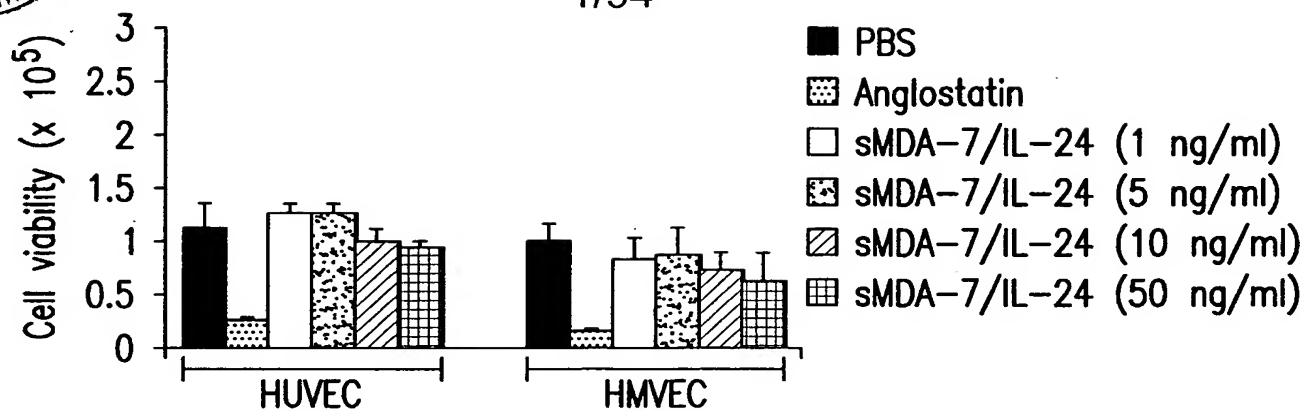


FIG. 1A

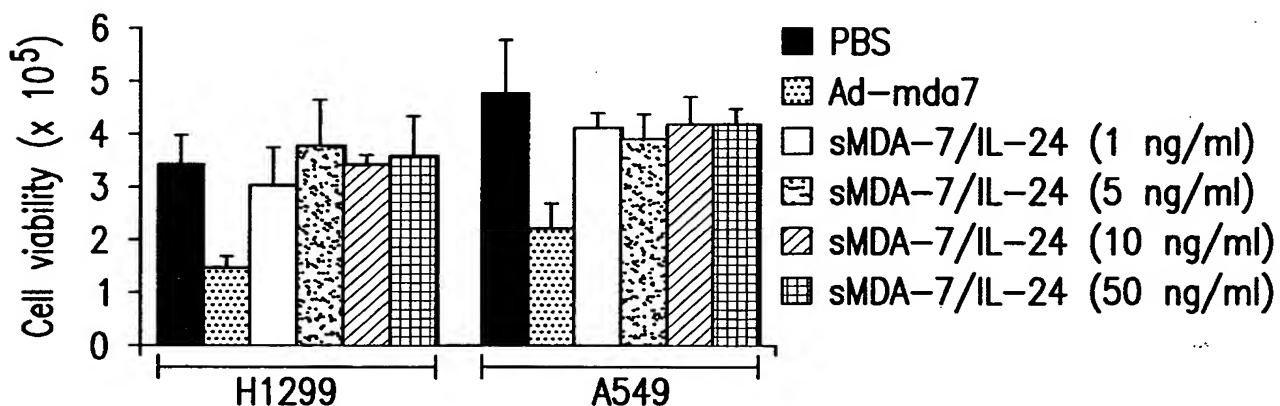


FIG. 1B

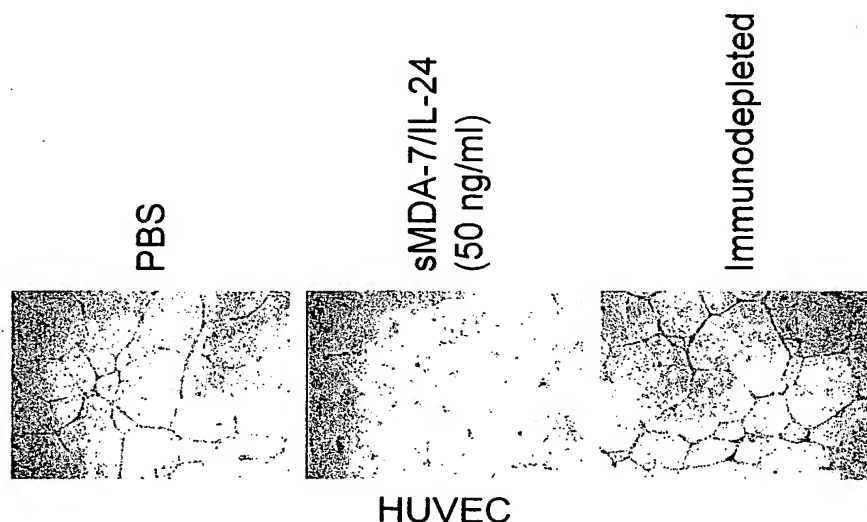


FIG. 1C

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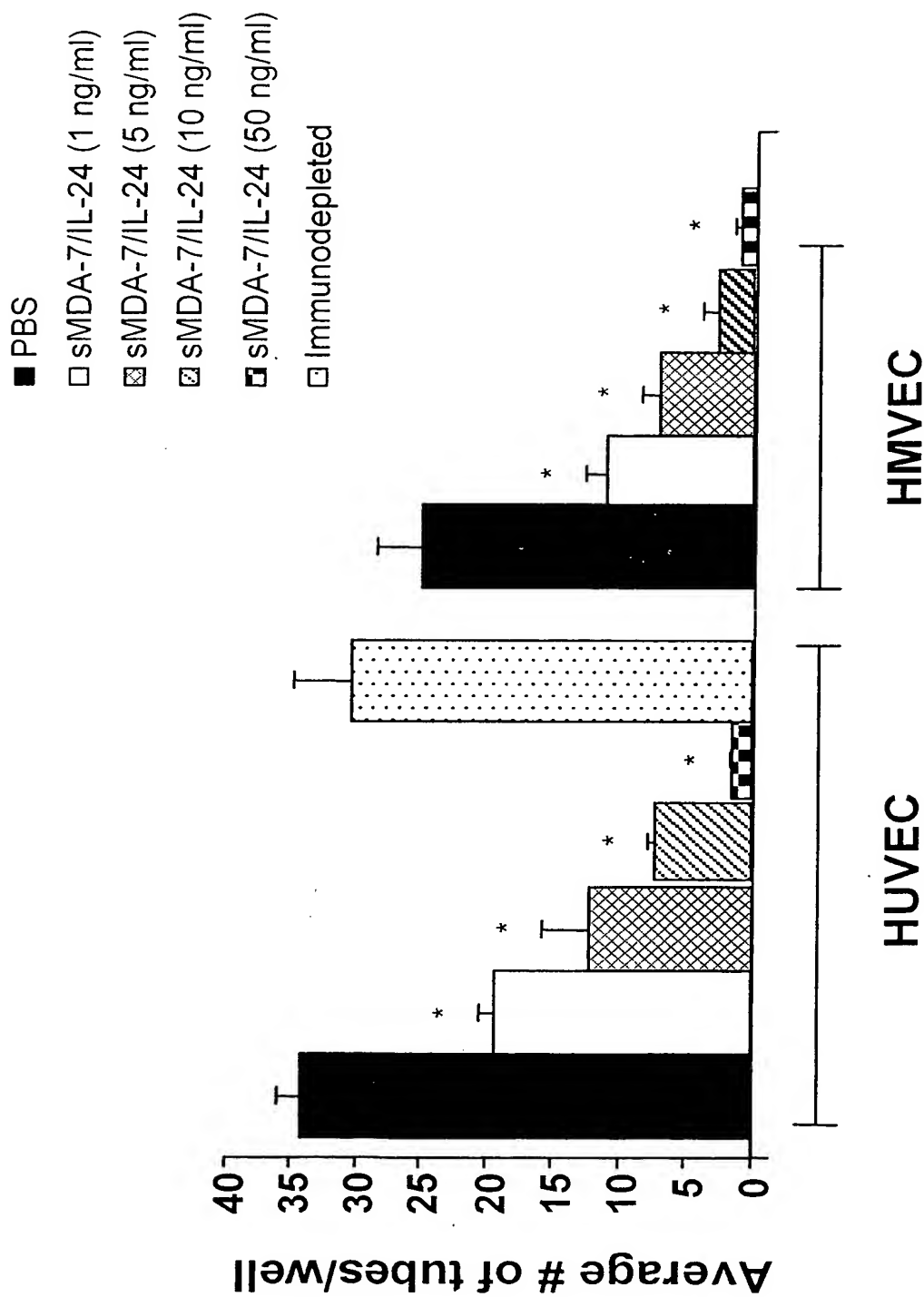
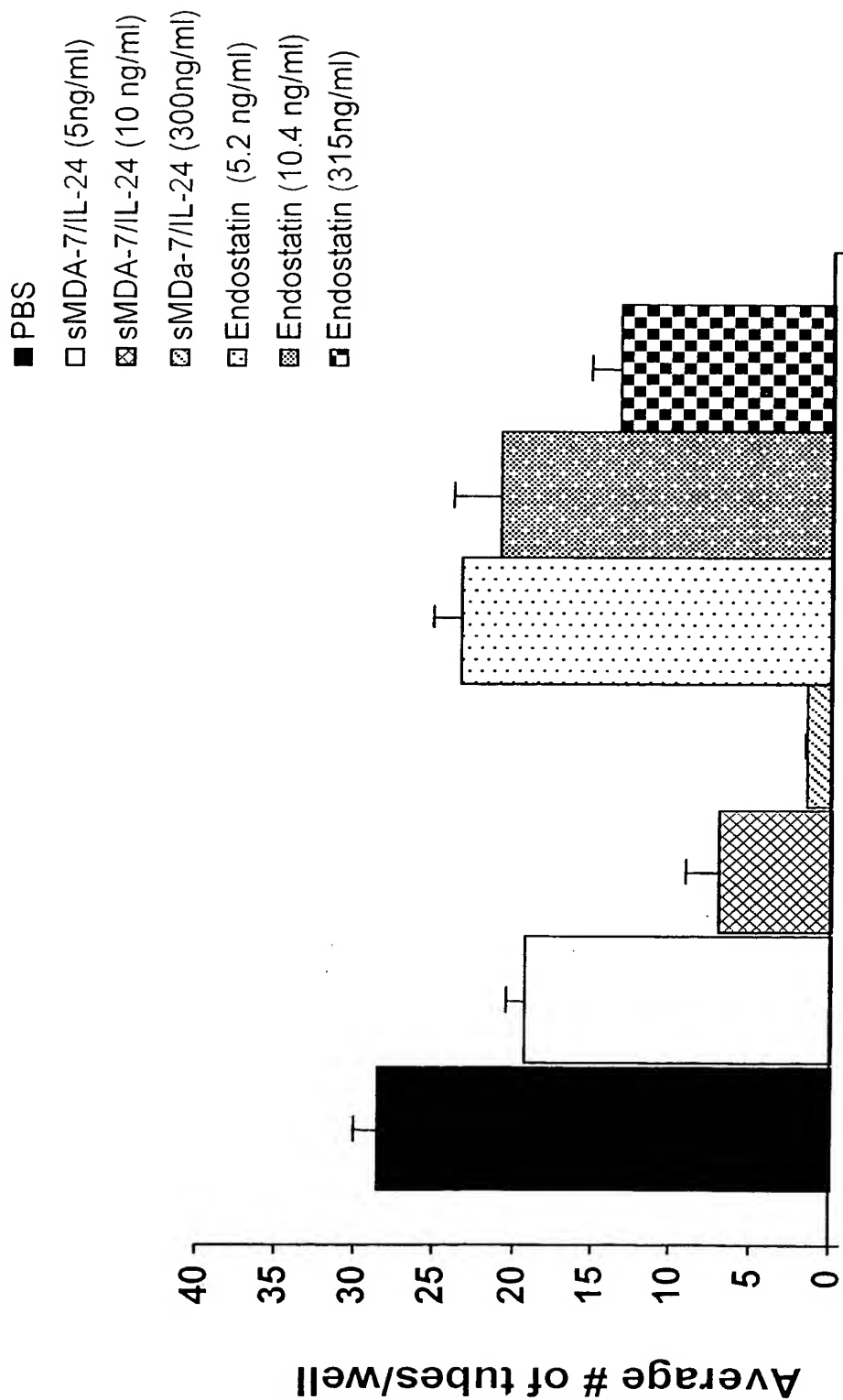


FIG.1D

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HUVEC

FIG.2

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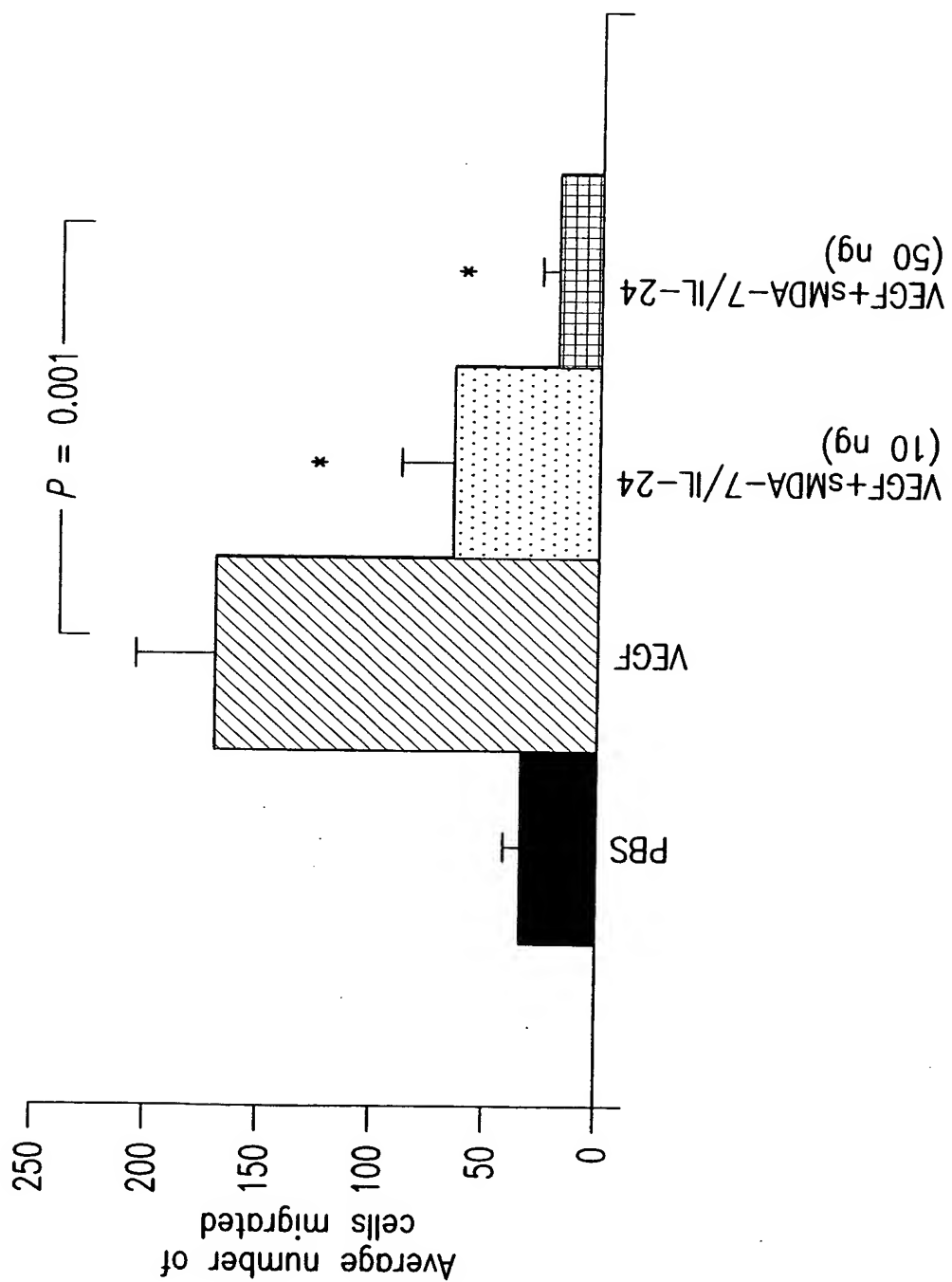
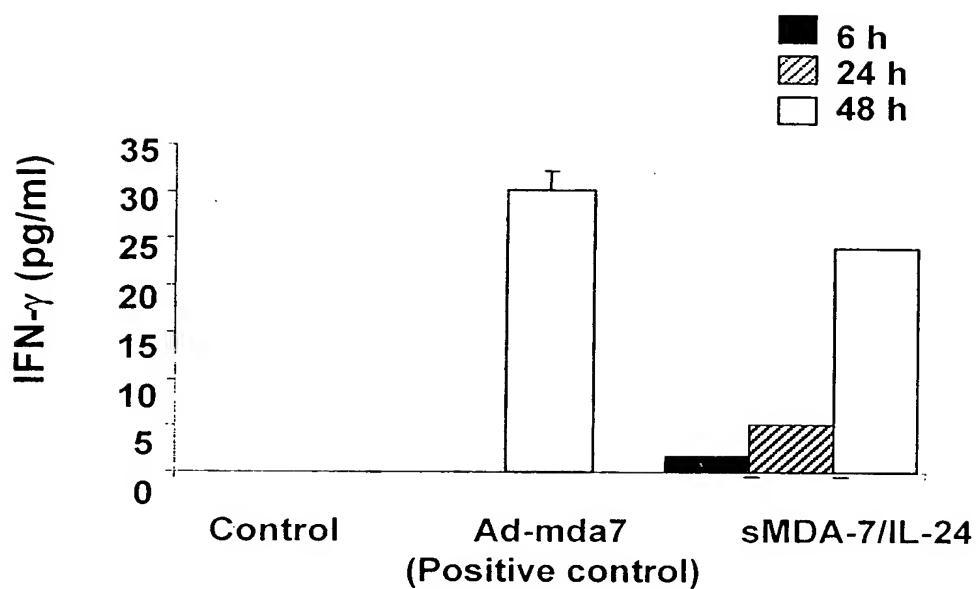


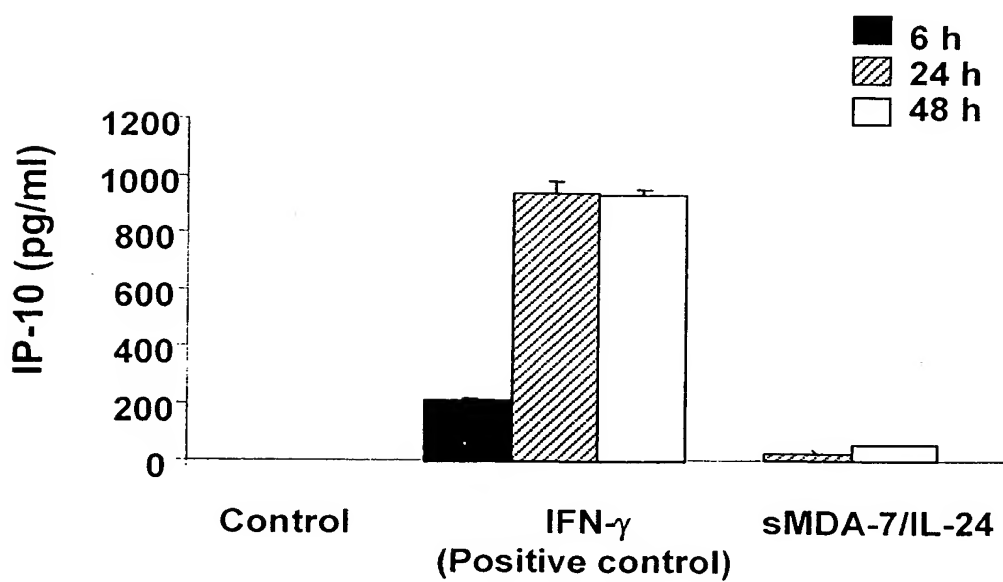
FIG. 3

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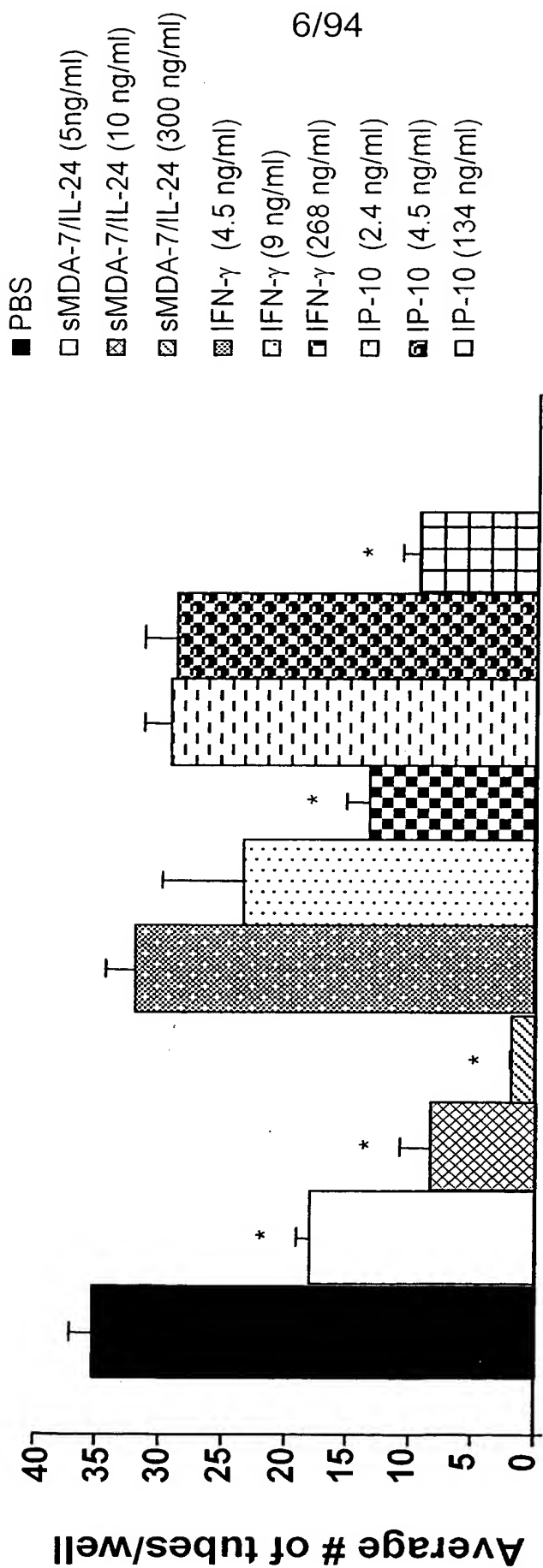
IFN- $\gamma$  assay

FIG.4A



IP-10 assay

FIG.4B



HUVEC

FIG.4C

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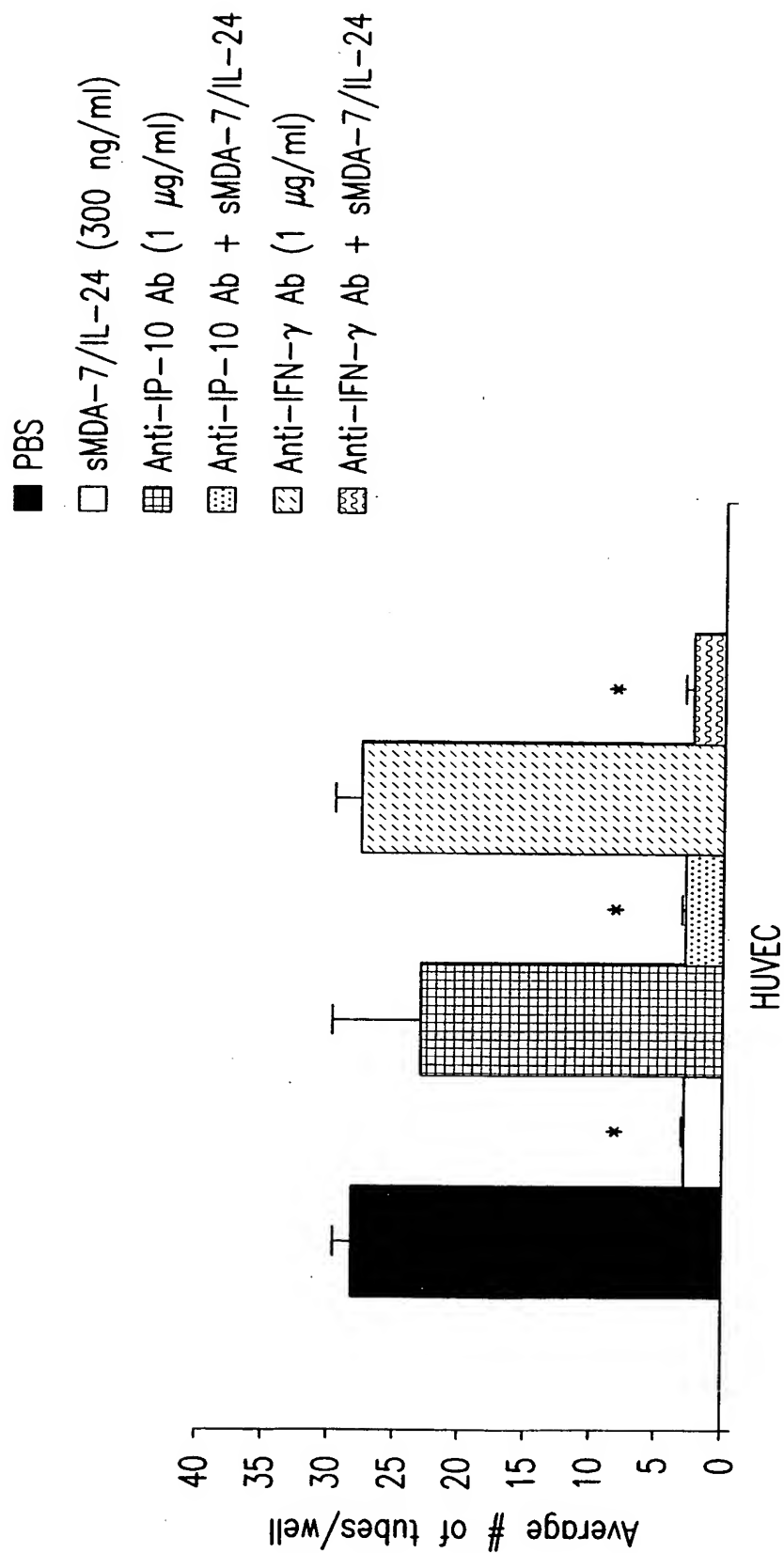


FIG.4D

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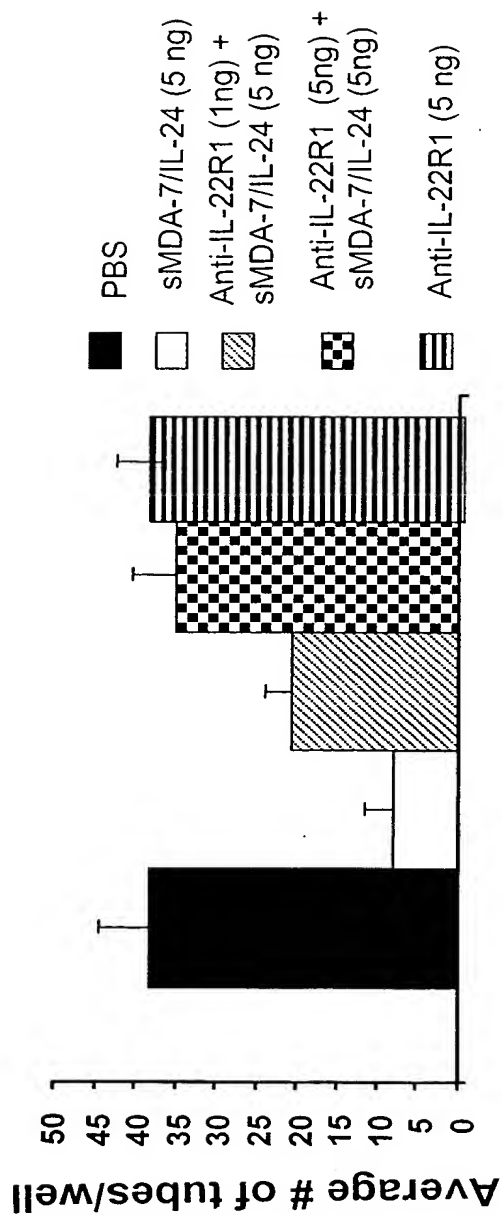


FIG. 5A

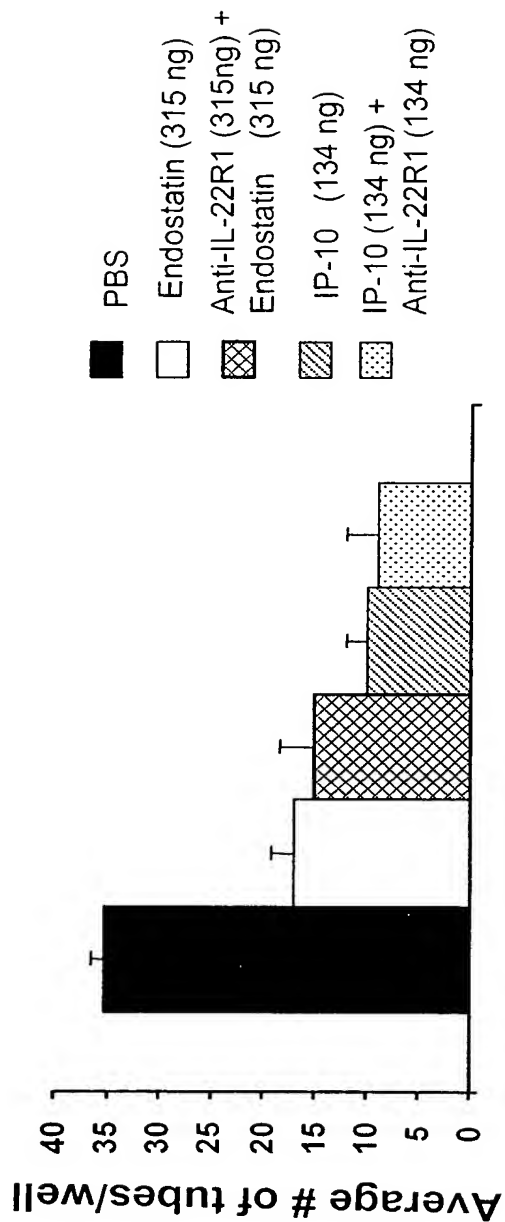


FIG. 5B



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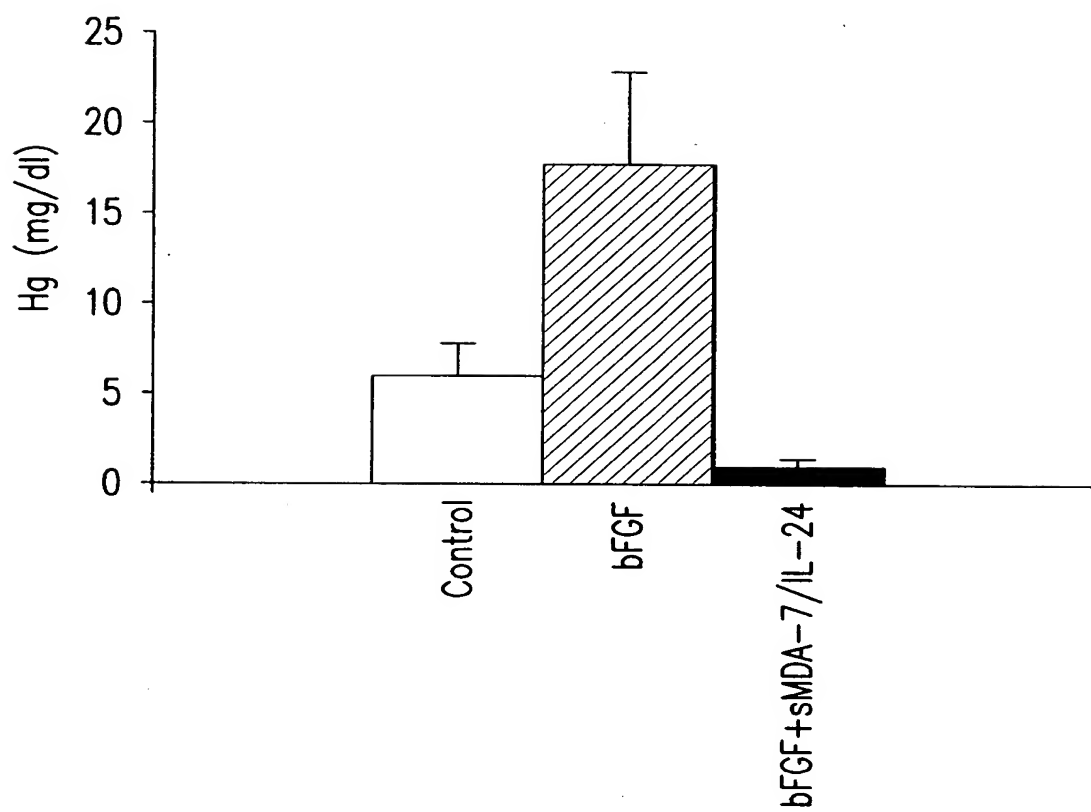


FIG.6A

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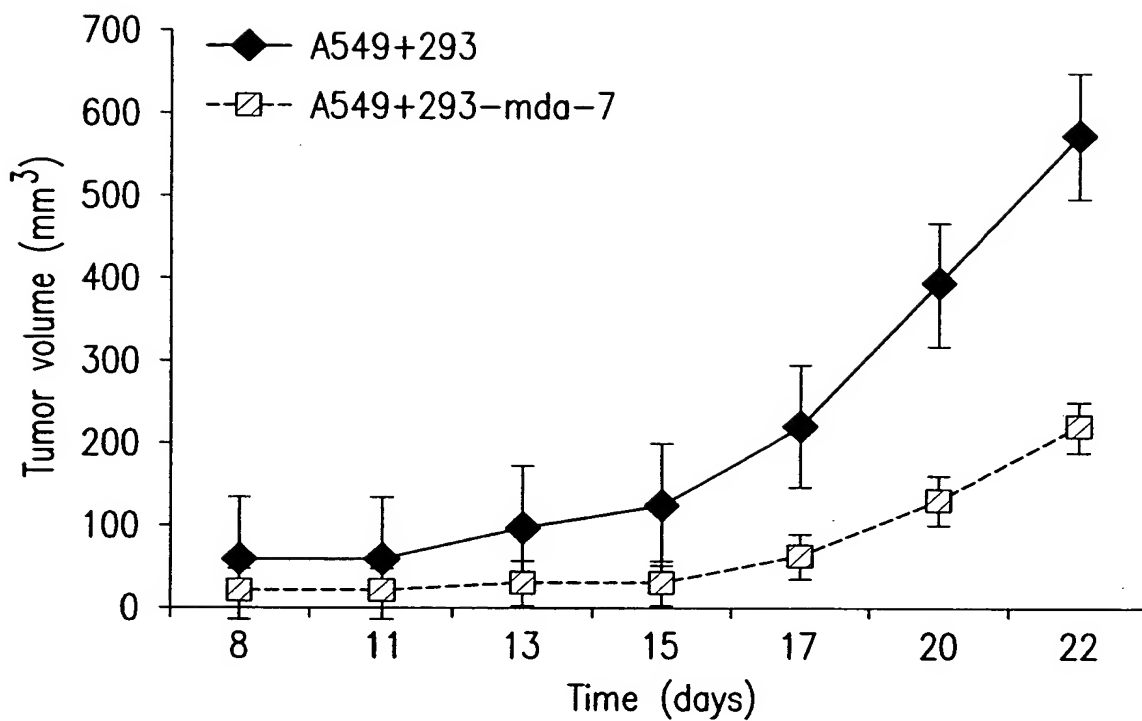


FIG.6B

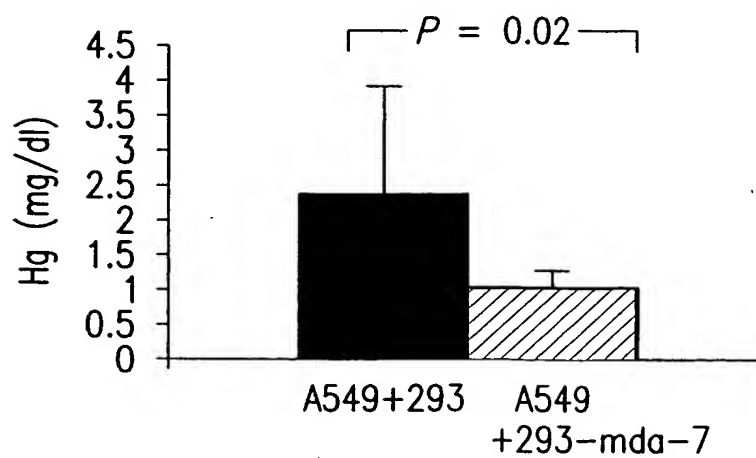


FIG.6C

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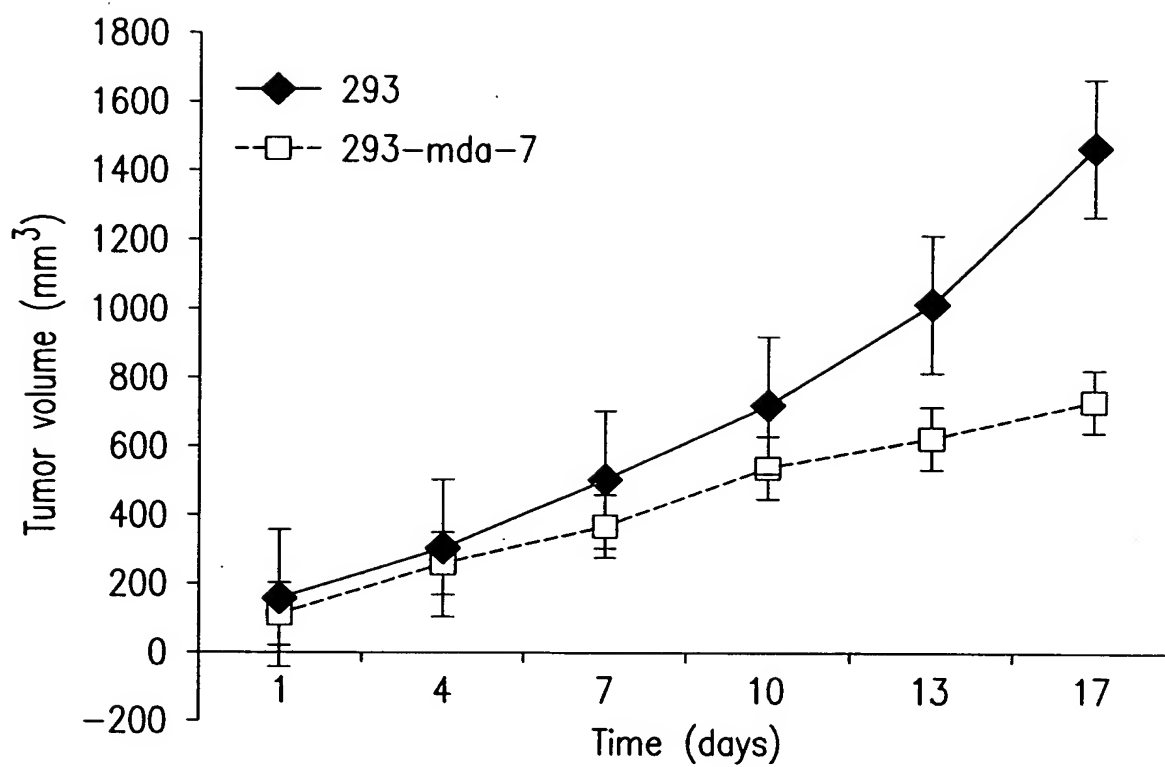


FIG.6D

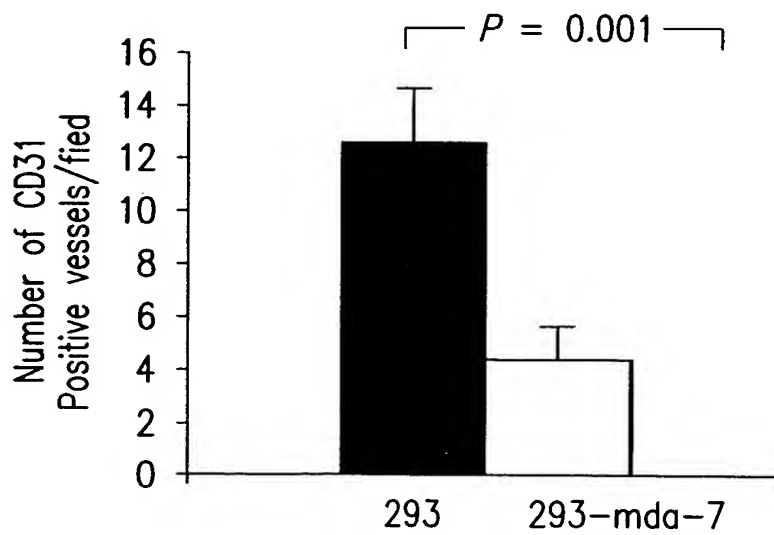


FIG.6E

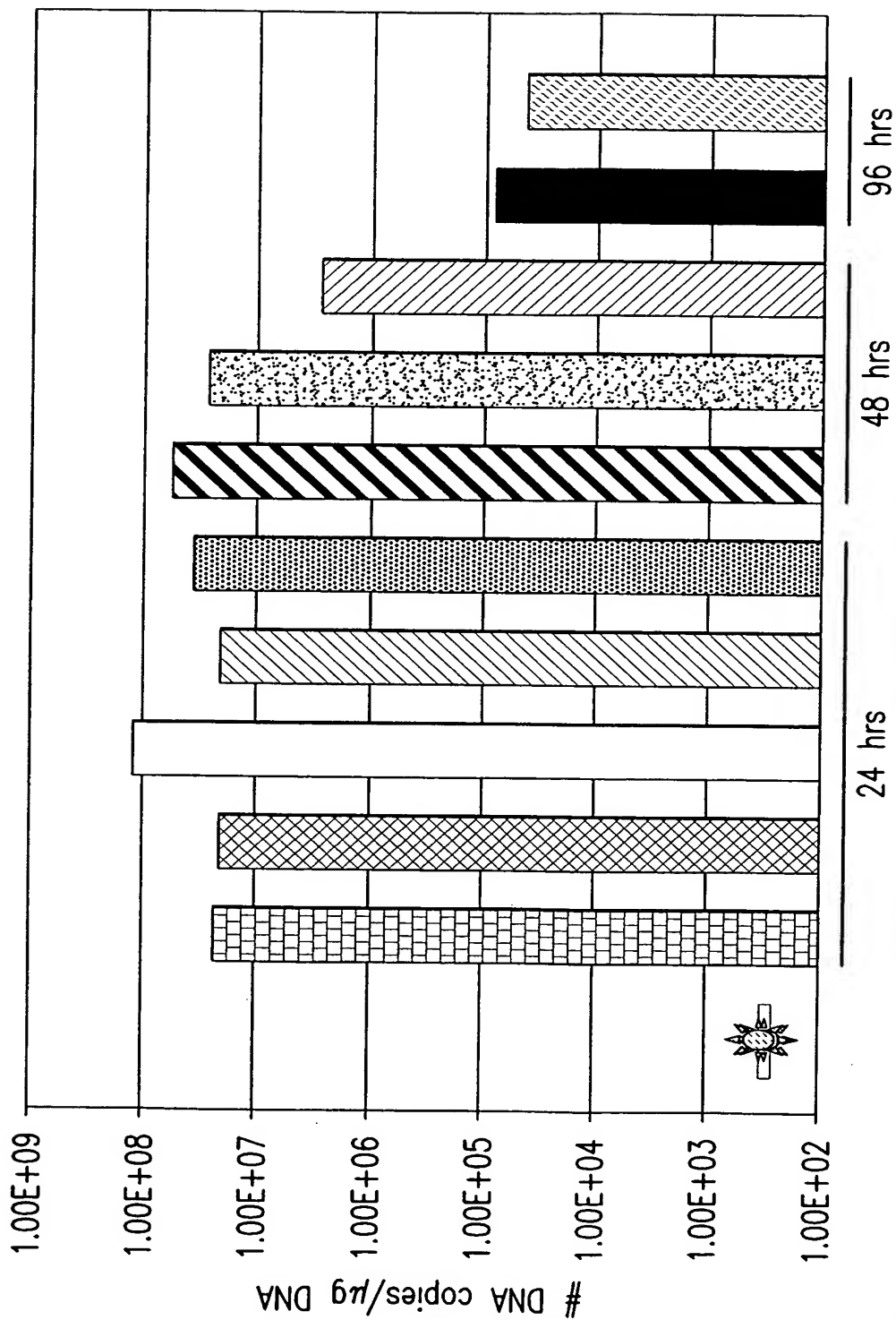
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## Study design

| Cohort # | # of patients | Viral dose<br>(particles)             | biopsy time<br>(post inj.) |
|----------|---------------|---------------------------------------|----------------------------|
| 1        | 1             | $2 \times 10^{10}$                    | 24 hrs                     |
| 2        | 1             | $2 \times 10^{11}$                    | 24 hrs                     |
| 3        | 3             | $2 \times 10^{12}$                    | 24 hrs                     |
| 4        | 3             | $2 \times 10^{12}$                    | 48 hrs                     |
| 5        | 3             | $2 \times 10^{12}$                    | 72/96 hrs                  |
| 6        | 3             | $2 \times 10^{12}$<br>(divided doses) | 48 hrs                     |
| 7        | 5             | $2 \times 10^{12}$                    | Core bx @30 days           |
| 8        | 5             | $2 \times 10^{12}$<br>(2x/wk x 3 wks) | Core bx @ 30 days          |

FIG.7

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⊙: Average pre-treat signal  
is 4.4E+02, n=8

FIG.8

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| Pt | Time (hr) | (Pre-treat MDA-7 IHC – all negative) |              |           |              |
|----|-----------|--------------------------------------|--------------|-----------|--------------|
|    |           | MDA-7 ctr                            | MDA-7 periph | TUNEL ctr | TUNEL periph |
| 1  | 24        | 20%                                  | 0%           | 20%       | 0%           |
| 2  | 24        | 30%                                  | 5%           | 70%       | 10%          |
| 3  | 24        | 75%                                  | 40%          | 50%       | 30%          |
| 4  | 24        | 35%                                  | 5%           | n.d.      | n.d.         |
| 5  | 24        | 50%                                  | 25%          | 25%       | 17%          |
| 6  | 48        | >60%                                 | 20%          | 40%       | 17%          |
| 7  | 48        | 60%                                  | 5%           | 70%       | 10%          |
| 8  | 48        | 20%                                  | 0%           | 5%        | 0%           |
| 9  | 96        | 50%                                  | 25%          | 80%       | 25%          |
| 10 | 96        | 90%                                  | 0%           | 35%       | 0%           |

MDA-7/TUNEL correlation  $p < 0.01$

FIG.9

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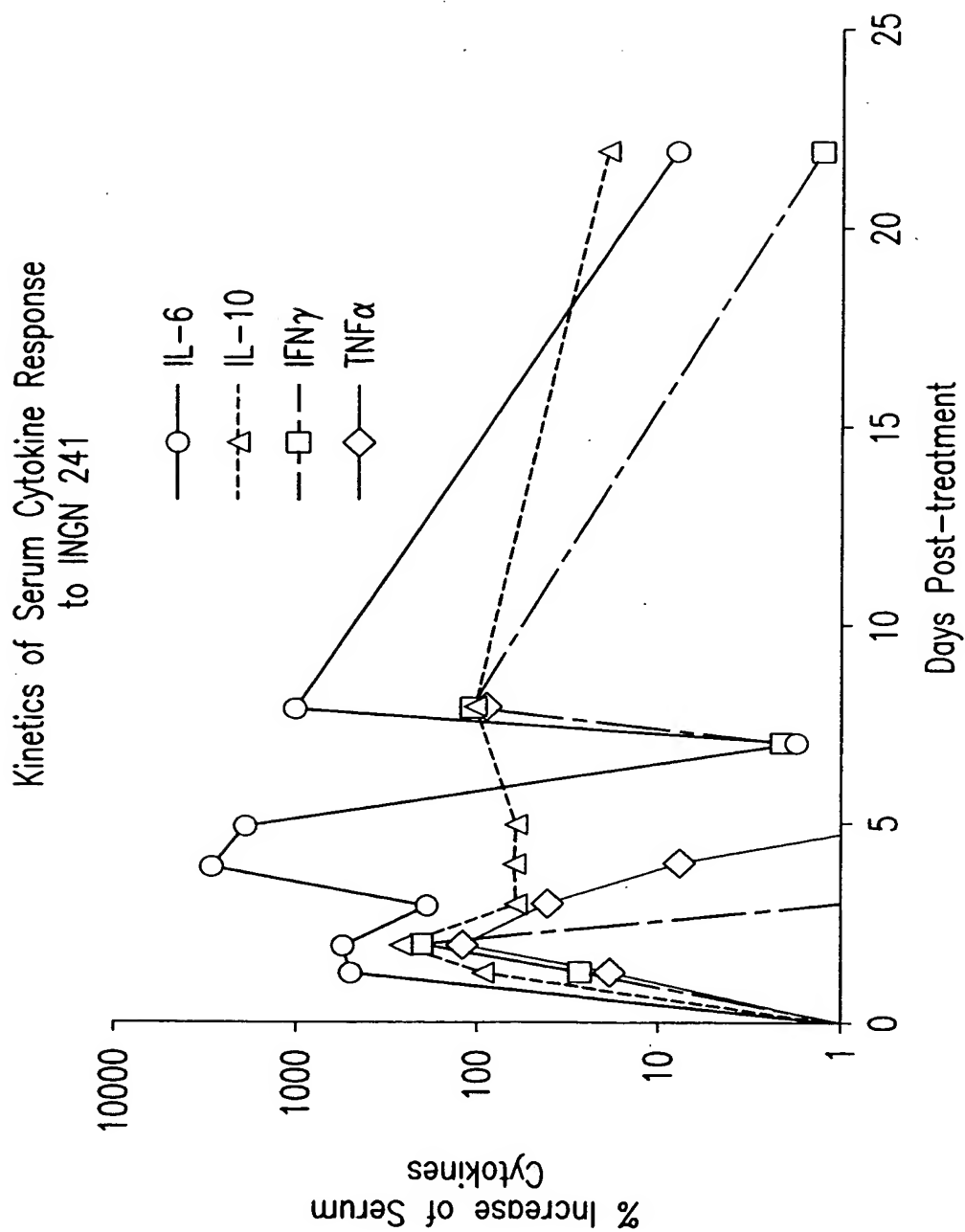


FIG.10

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Serum Cytokine Response to  
Intratumoral INGN 241 Treatment.

| Cohort<br><u>(dose)</u>     | No. pts<br><u>tested</u> | <u>No. positive (mean % peak increase)</u> |              |                               |                               |               |
|-----------------------------|--------------------------|--|--------------|-------------------------------|-------------------------------|---------------|
|                             |                          | <u>IL-6</u>                                | <u>IL-10</u> | <u>IFN<math>\gamma</math></u> | <u>TNF<math>\alpha</math></u> | <u>GM-CSF</u> |
| 1<br>( $2 \times 10^{10}$ ) | 1                        | 1<br>(298%)                                | 1<br>(291%)  | 1<br>(>1,000%)                | 0                             | 1 (150%)      |
| 2<br>( $2 \times 10^{11}$ ) | 1                        | 1<br>(143%)                                | 1<br>(599%)  | 1<br>(281%)                   | 1<br>(864%)                   | 0             |
| 3<br>( $2 \times 10^{12}$ ) | 3                        | 1<br>(>1,000%)                             | 1<br>(134%)  | 1<br>(317%)                   | 2<br>(345%)                   | 1<br>(100%)   |
| 4<br>( $2 \times 10^{12}$ ) | 3                        | 3<br>(>1,000%)                             | 3<br>(221%)  | 1<br>(173%)                   | 2<br>(71%)                    | 1<br>(120%)   |
| 5<br>( $2 \times 10^{12}$ ) | 3                        | 3<br>(640%)                                | 3<br>(400%)  | 1<br>(317%)                   | 2<br>(156%)                   | 0             |
| 7<br>( $2 \times 10^{12}$ ) | 7                        | 5<br>(604%)                                | 6<br>(387%)  | 3<br>(860%)                   | 3<br>(255%)                   | 0             |
| Total                       | 18                       | 14   | 15           | 8                             | 10                            | 3             |

FIG.11



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Level of Increased CD8 + T cell Frequency in Pts  
who Received Intratumoral INGN 241.

| Cohort<br>(dose)           | No. pts<br>tested | No. with<br>increased<br>CD8 + T cells | Mean % CD3 + CD8 + T cells <sup>1</sup> |                      |                    |
|----------------------------|-------------------|--|---|----------------------|--------------------|
|                            |                   |  | Pre-<br>treatment                       | Post-<br>treatment @ | Day<br>Post-treat. |
| 1<br>(2x10 <sup>10</sup> ) | 1                 | 0                                      | 29%                                     | 27%                  |                    |
| 2<br>(2x10 <sup>11</sup> ) | 1                 | 1                                      | 32%                                     | 44%                  | Day 15             |
| 3<br>(2x10 <sup>12</sup> ) | 3                 | 3                                      | 33%                                     | 59%                  | Day 15             |
| 4<br>(2x10 <sup>12</sup> ) | 3                 | 1                                      | 35%                                     | 44%                  | Day 30             |
| 5<br>(2x10 <sup>12</sup> ) | 3                 | 1                                      | 22%                                     | 29%                  | Day 15             |
| 7<br>(2x10 <sup>12</sup> ) | 7                 | 2                                      | 30%                                     | 40%                  | Days 15-30         |
| Total                      | 18                | 8                                      | 31%                                     | 44%                  | Day 15             |

FIG.12

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Increase in Peripheral Blood CD8 + T Cells  
Following intratumoral INGN 241 Injection

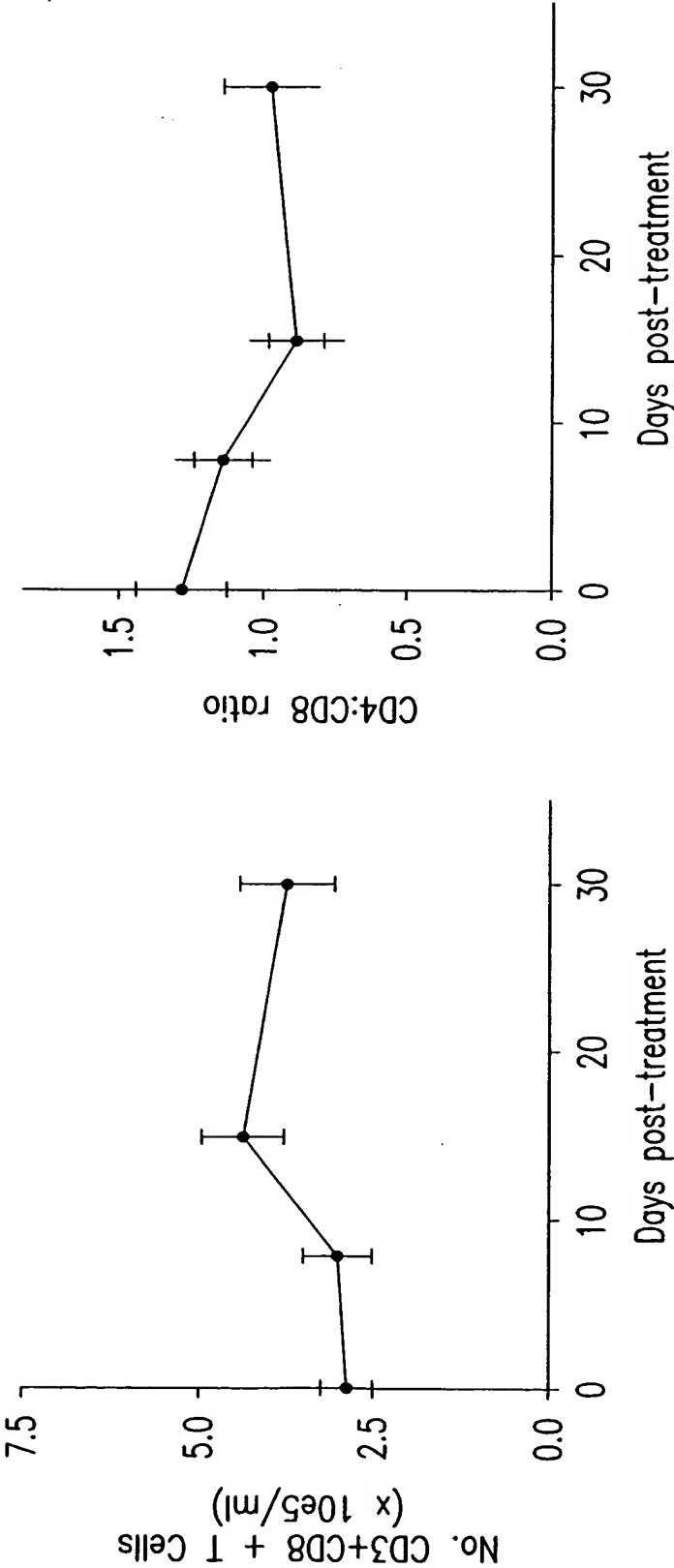


FIG.13

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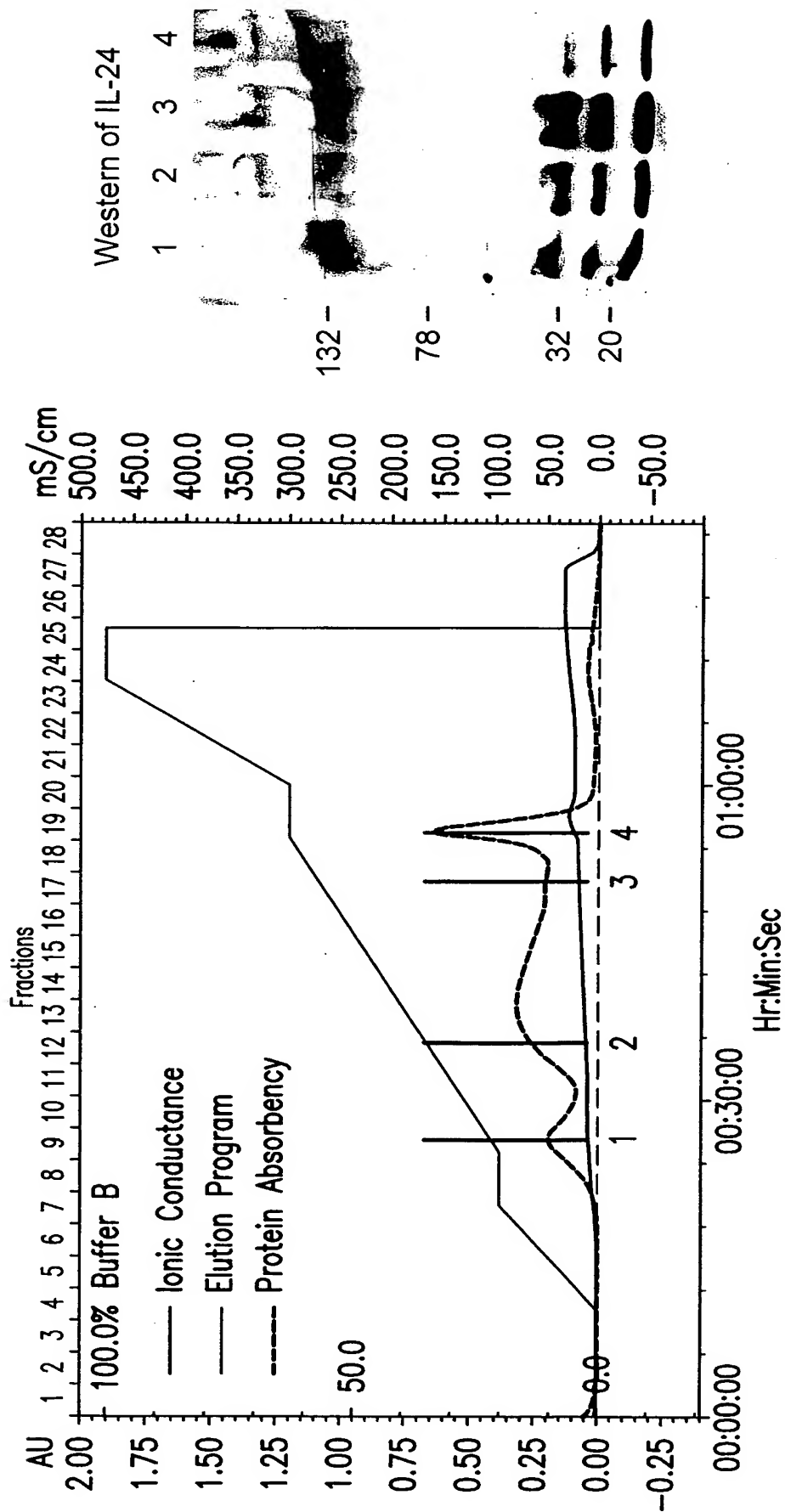


FIG.14

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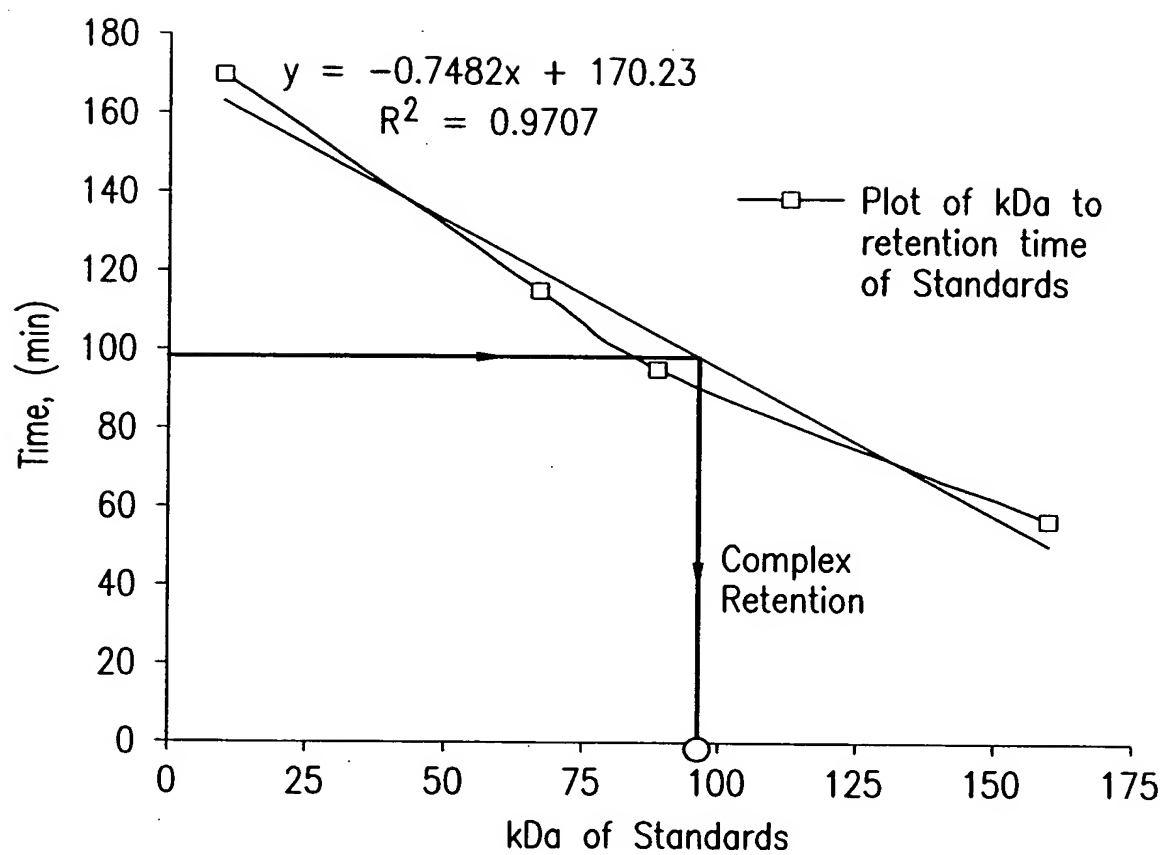
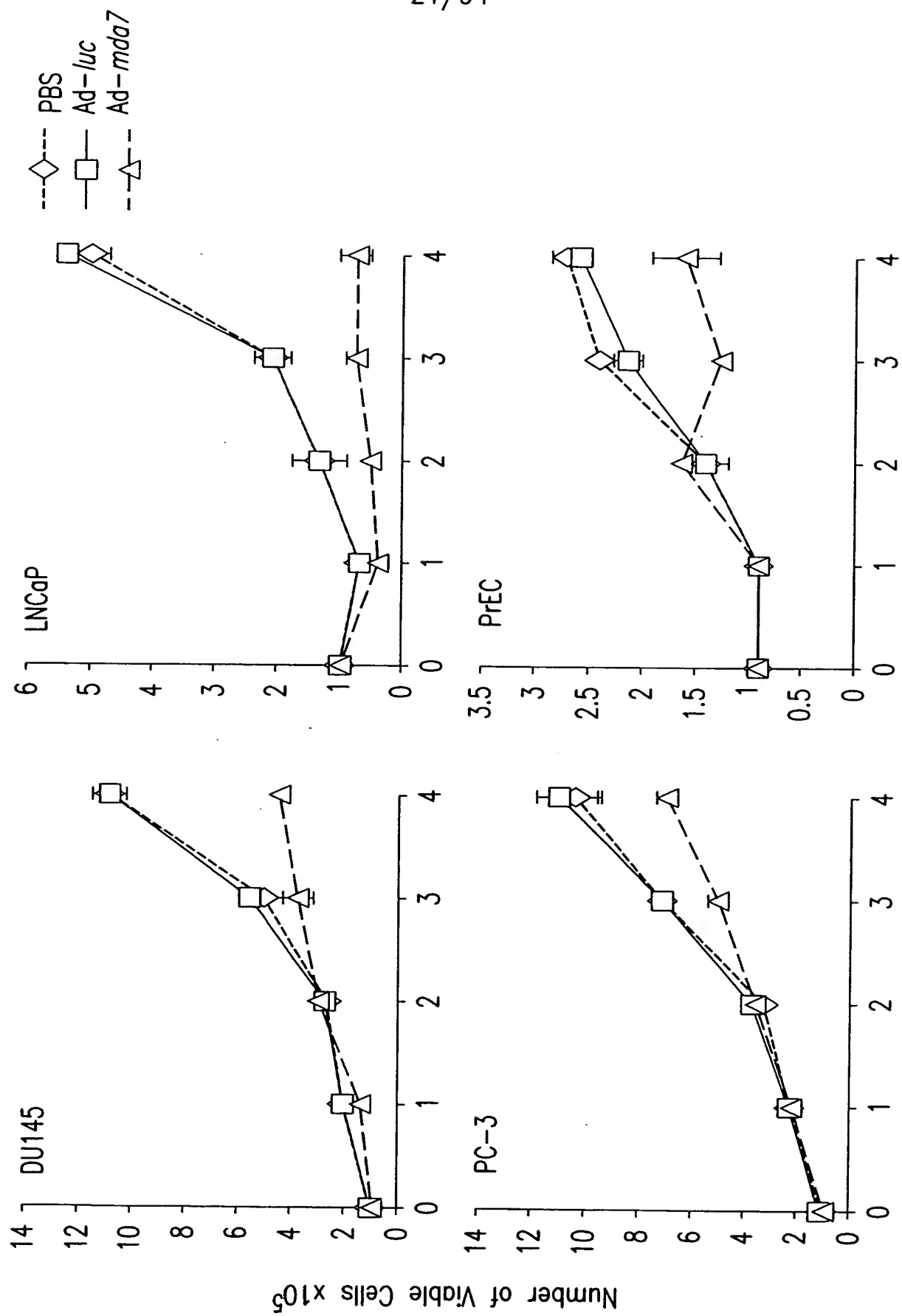


FIG.15

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Days after Infection

FIG.16

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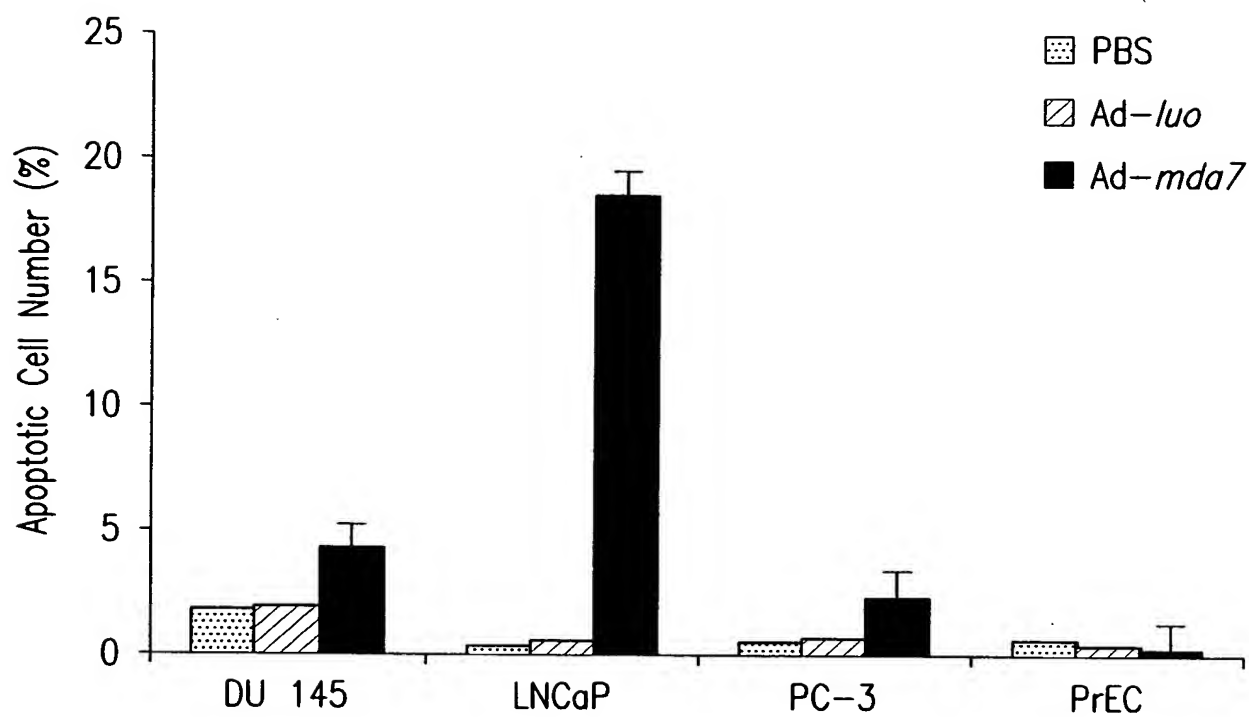


FIG.17

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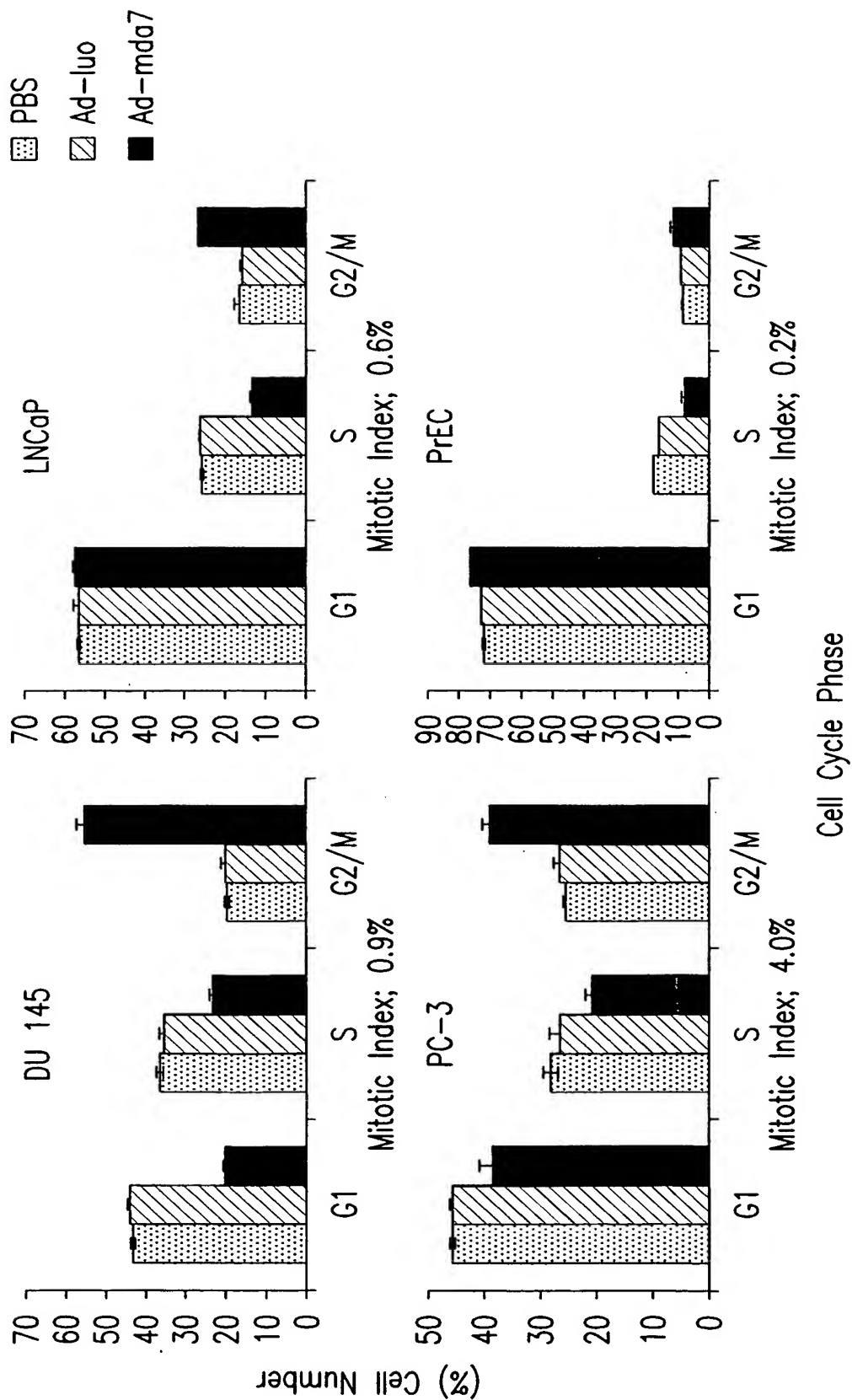


FIG.18

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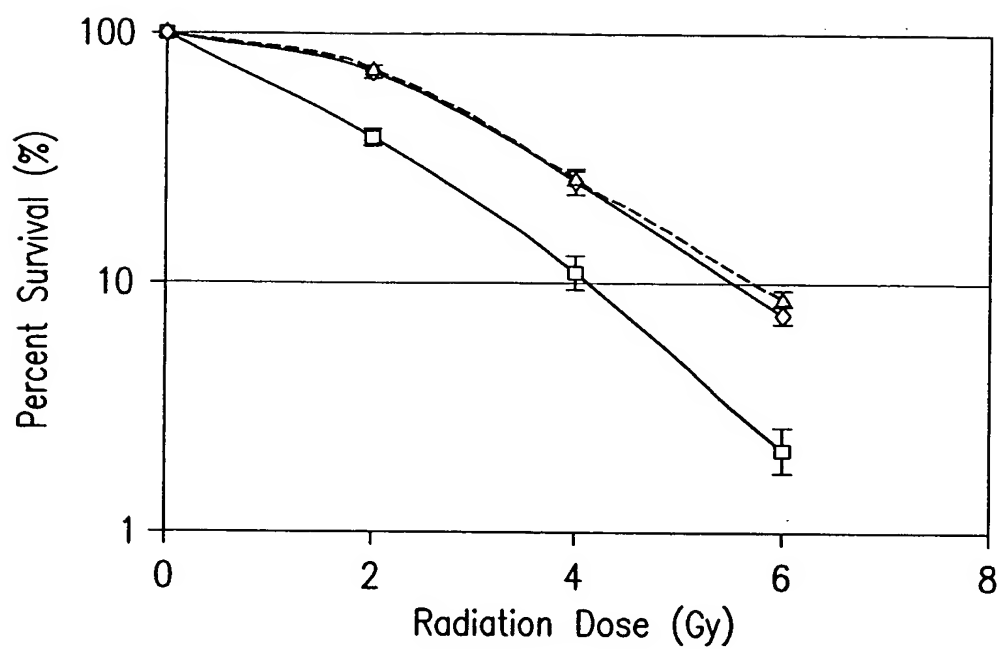


FIG.19A

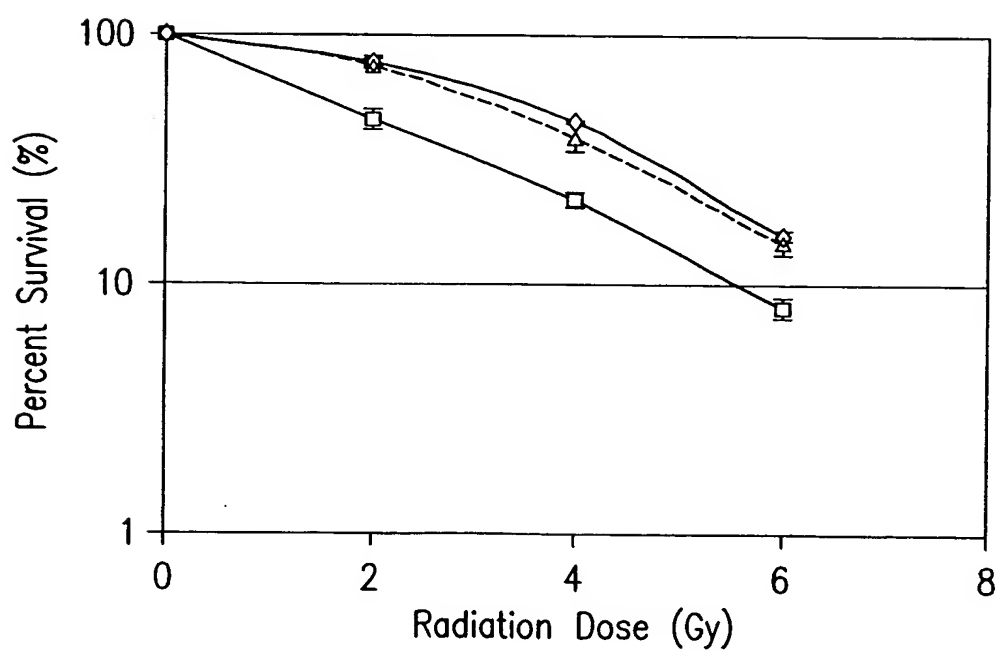


FIG.19B



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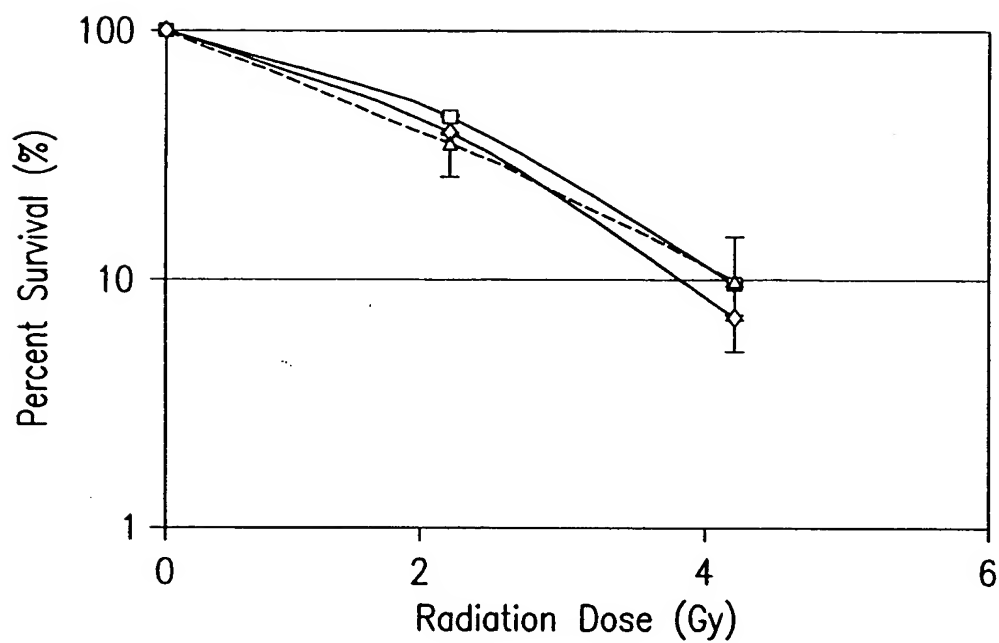


FIG.19C

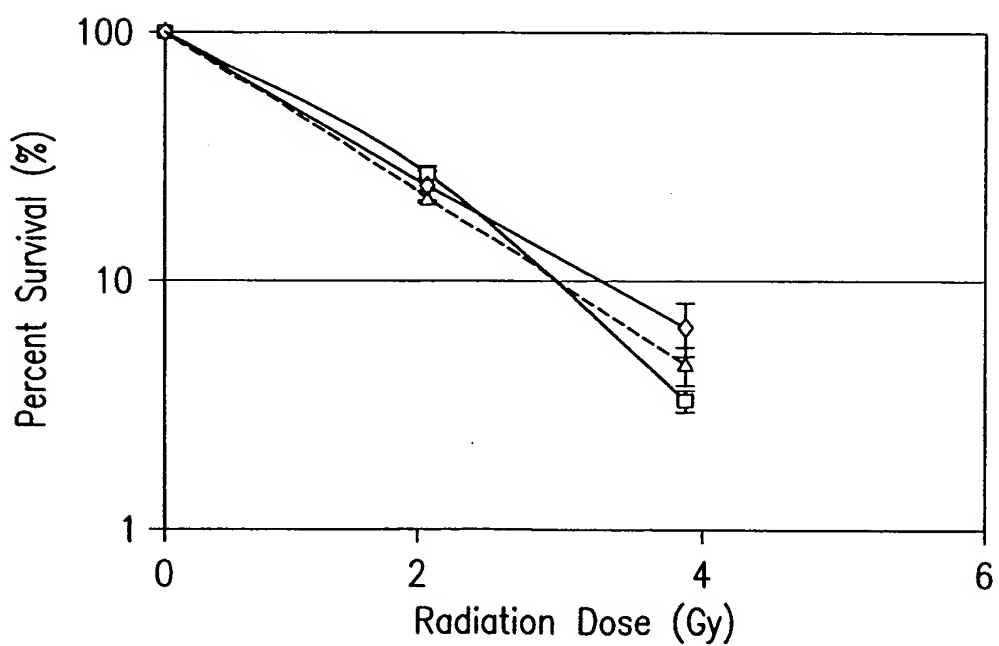


FIG.19D

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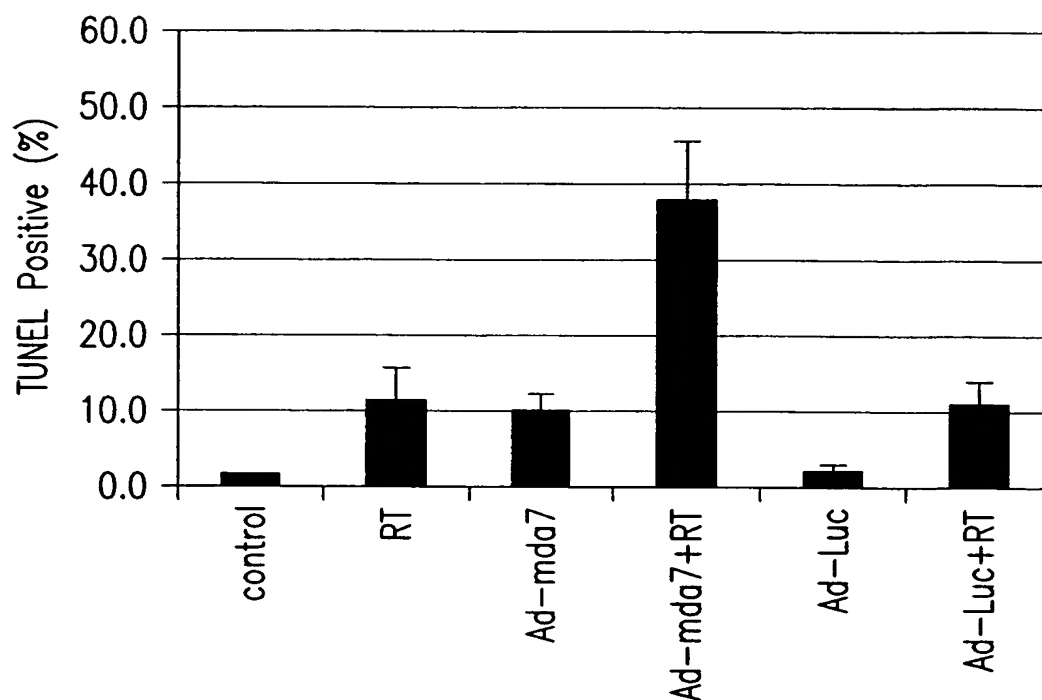


FIG. 20A

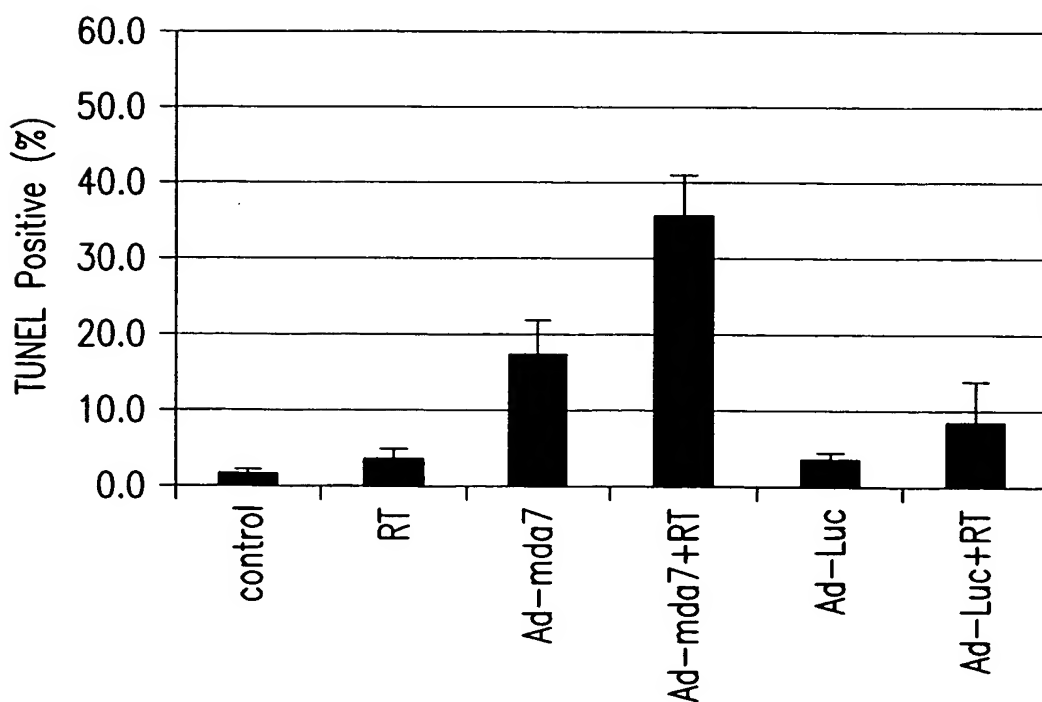


FIG. 20B

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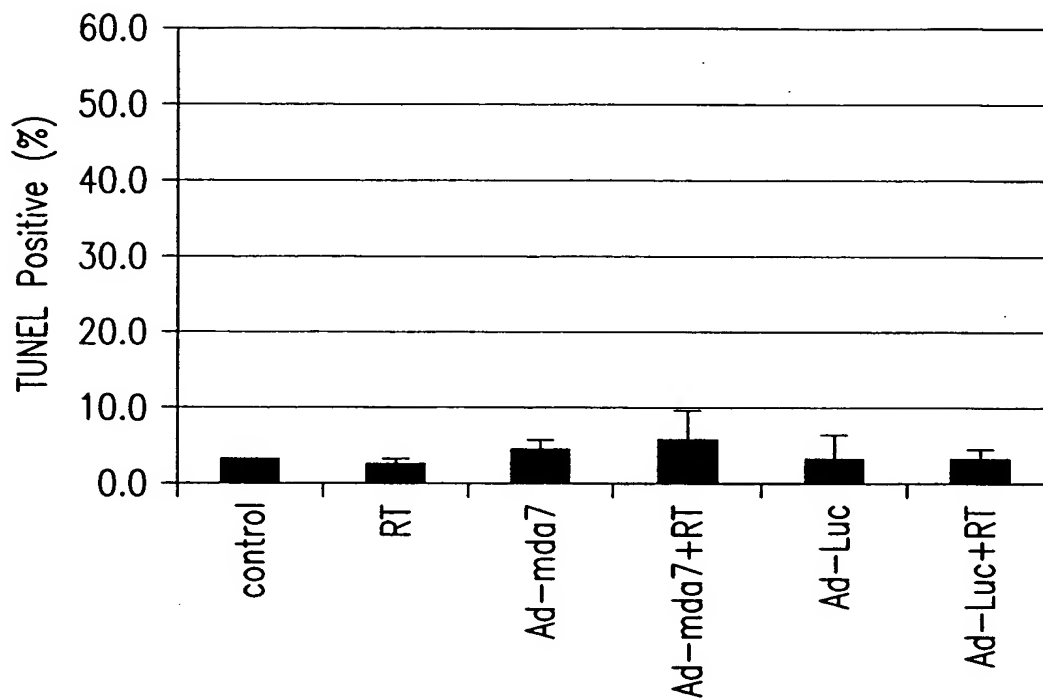


FIG.20C

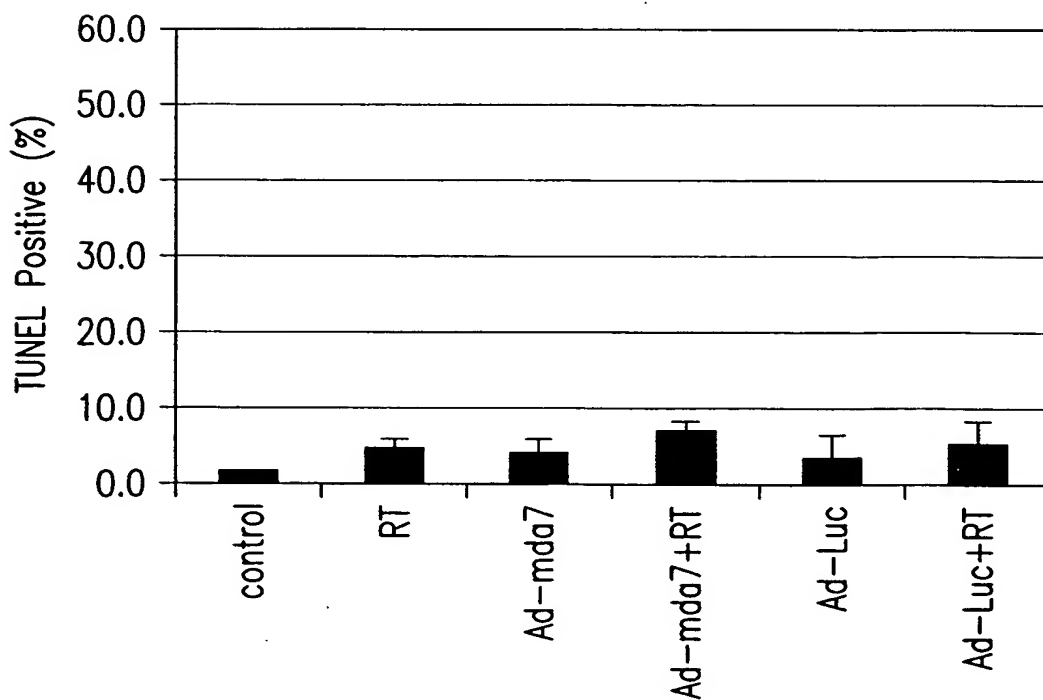
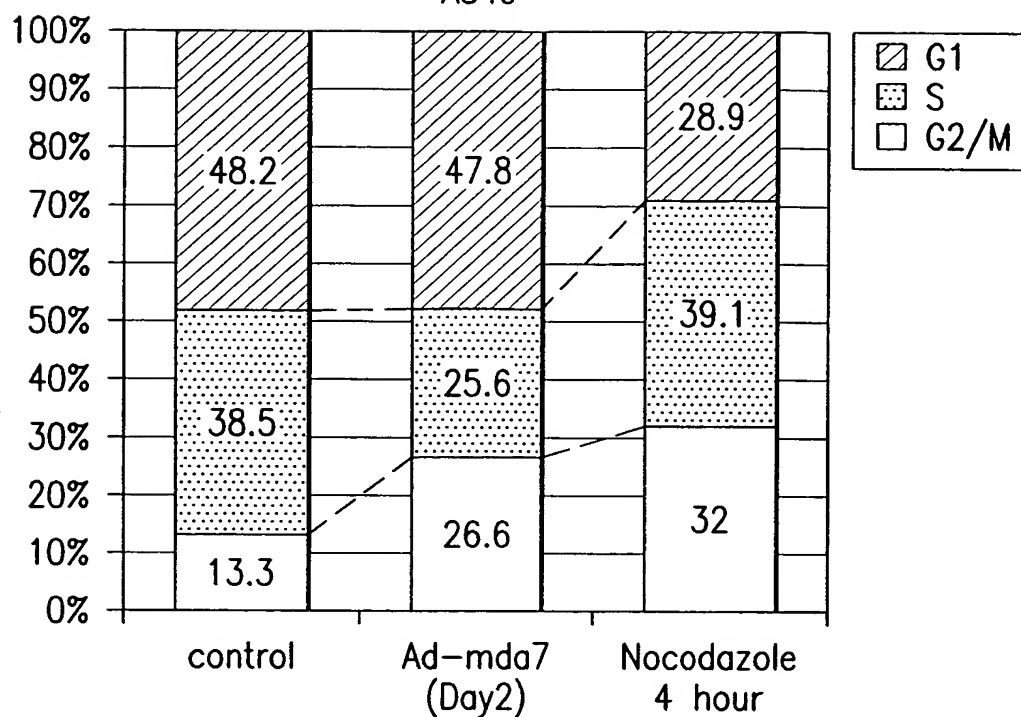


FIG.20D

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A549



H1299

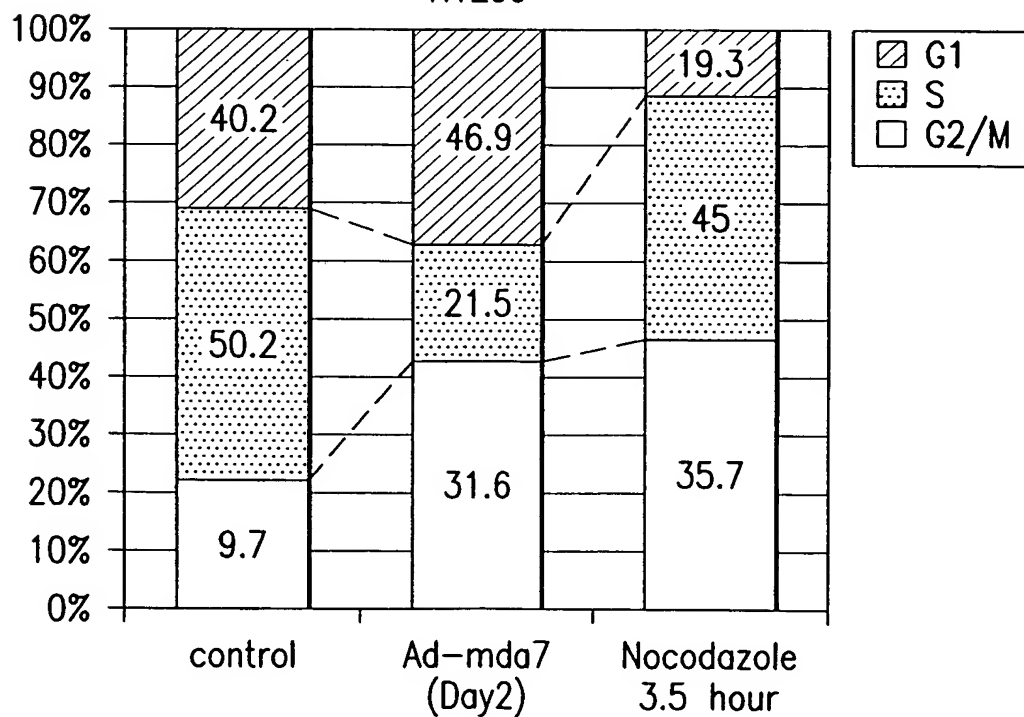


FIG.21

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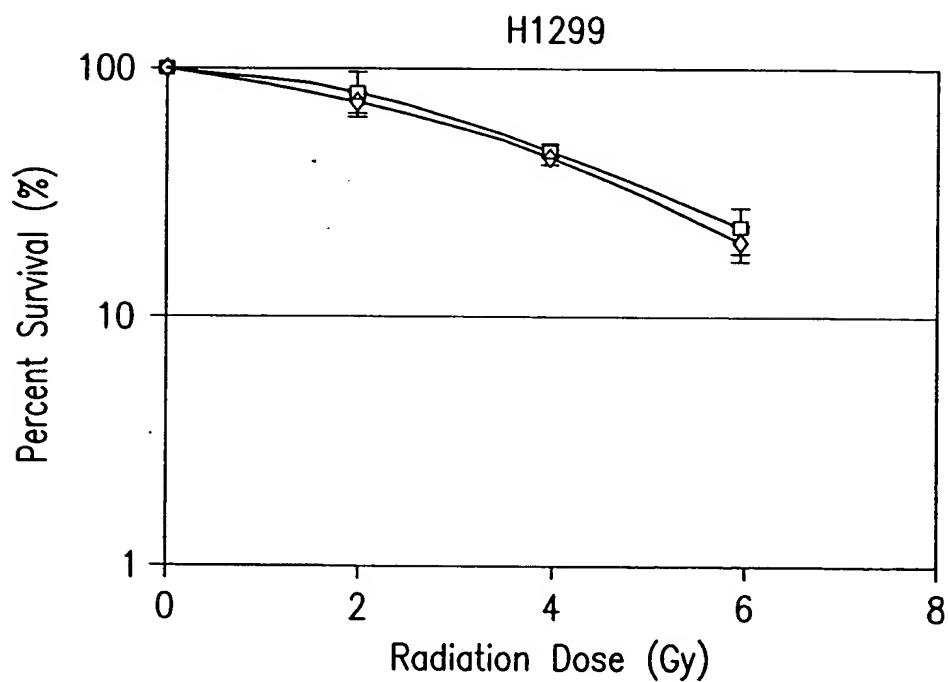
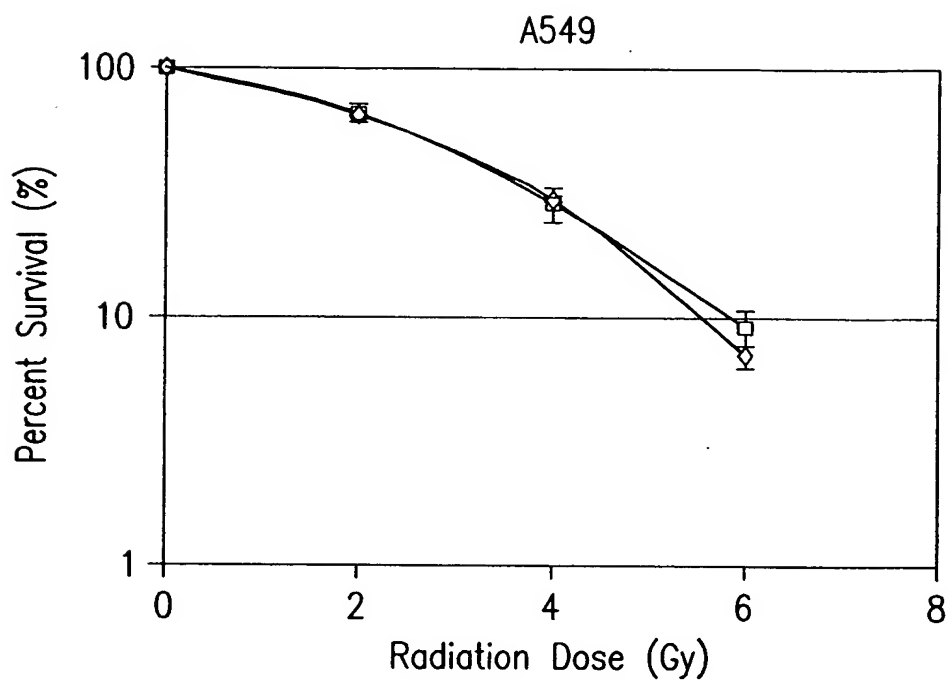


FIG.22

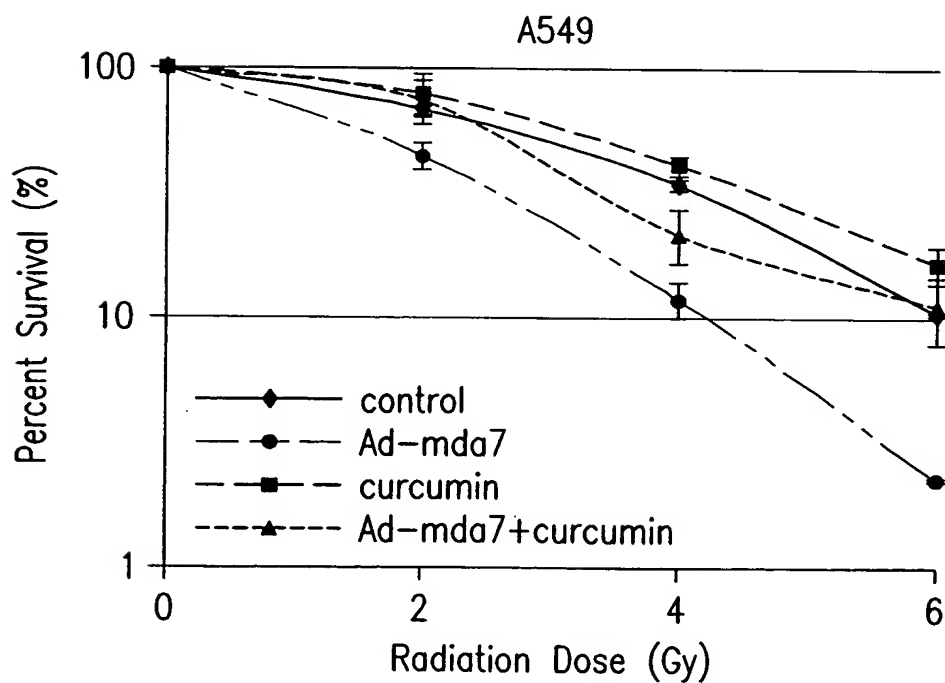
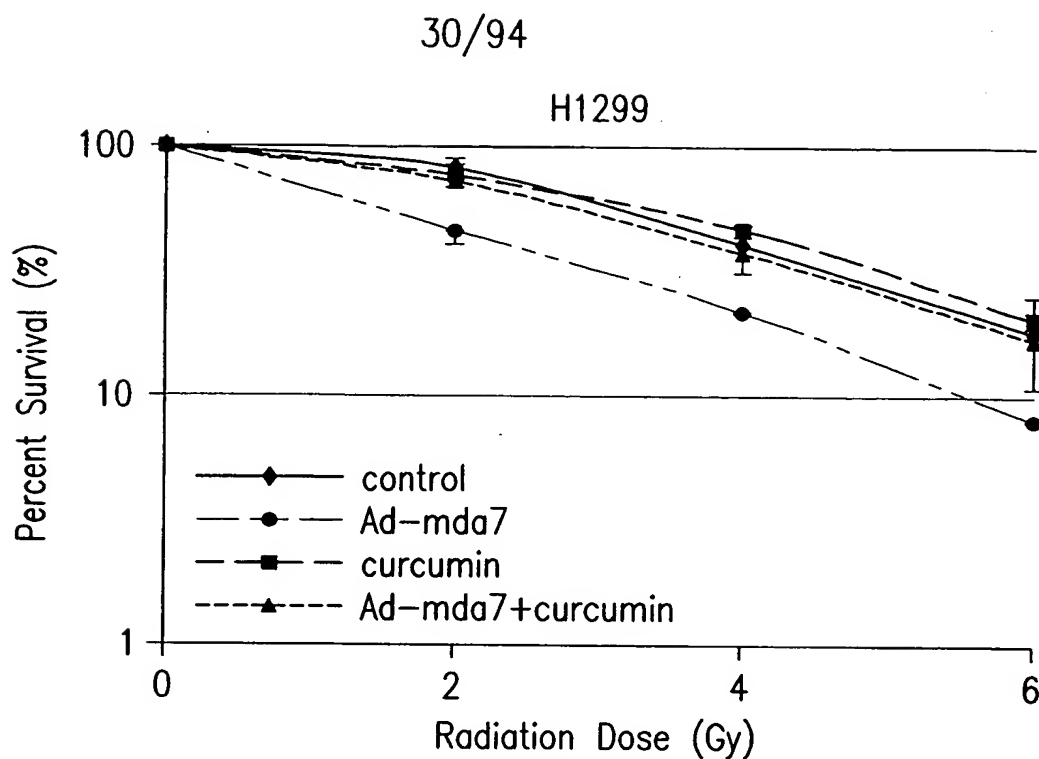


FIG.23

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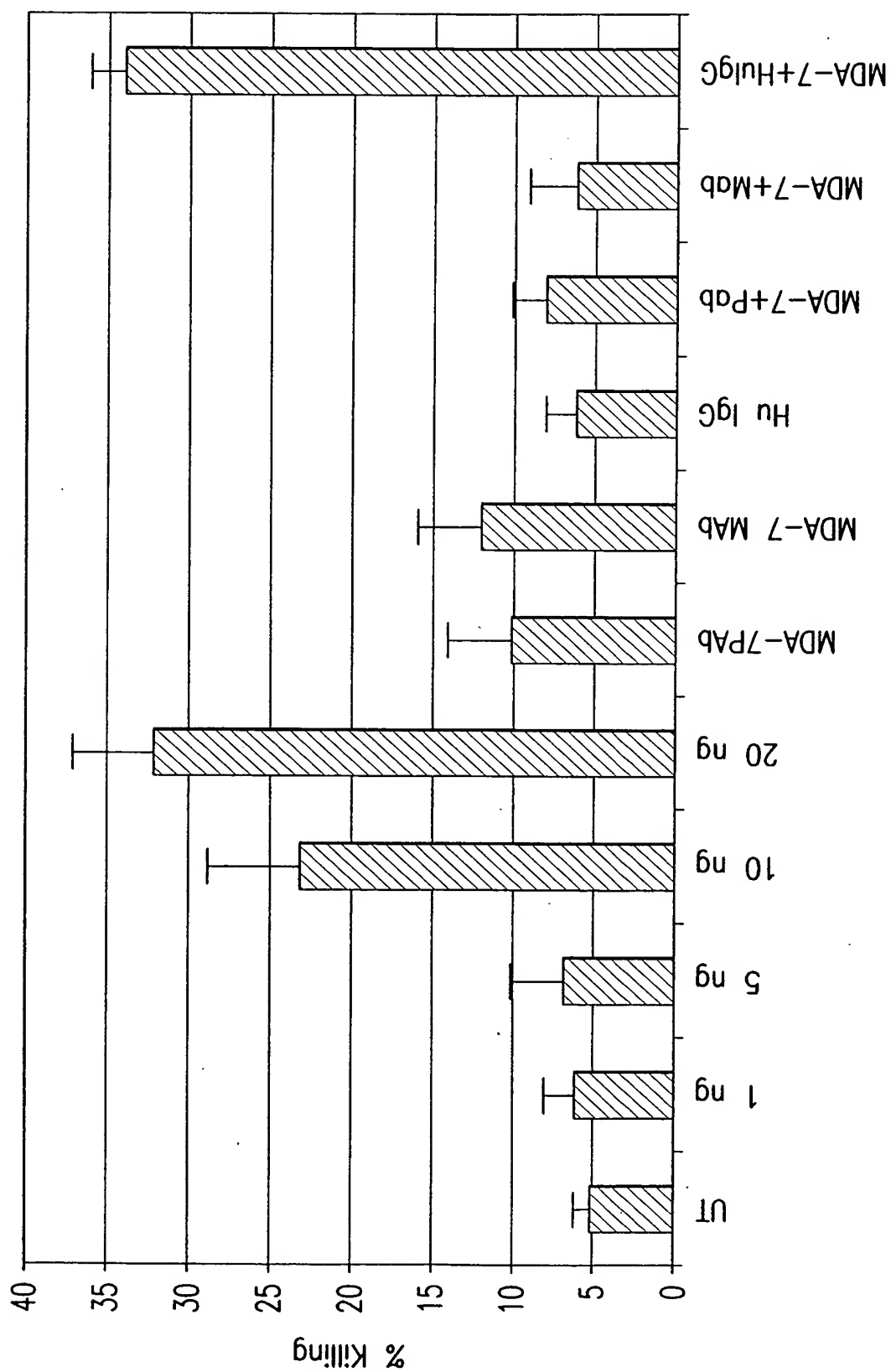


FIG.24

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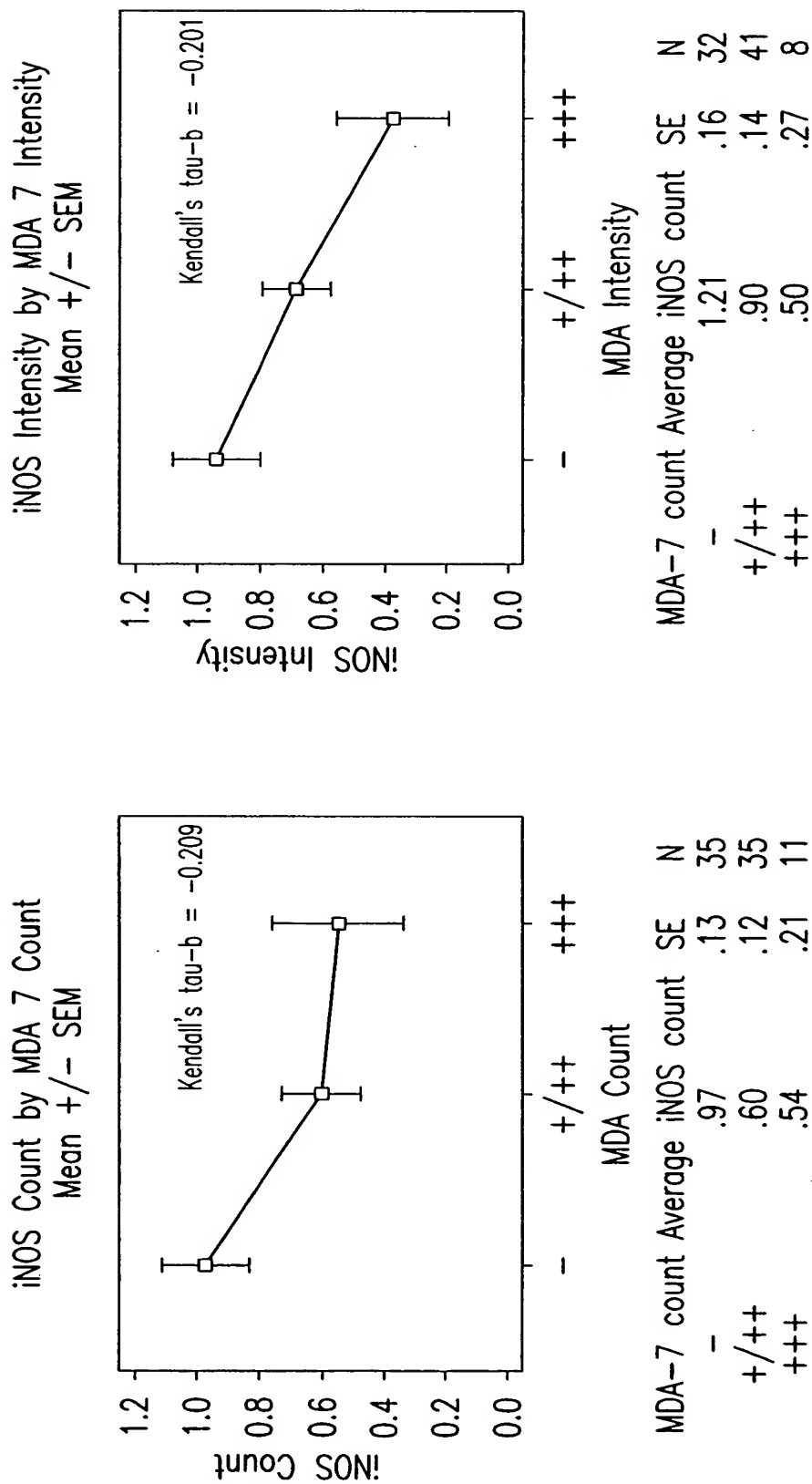


FIG.25B

FIG.25A



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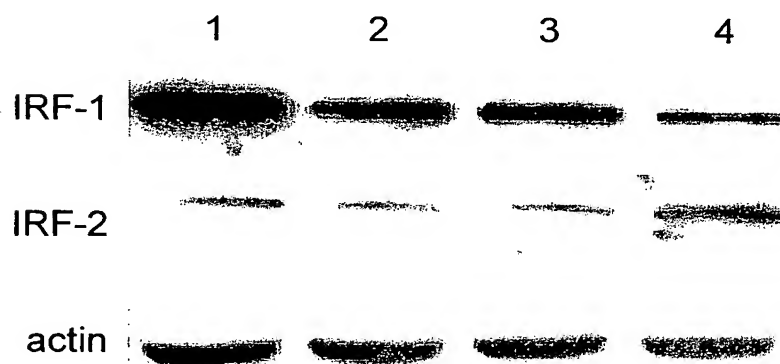
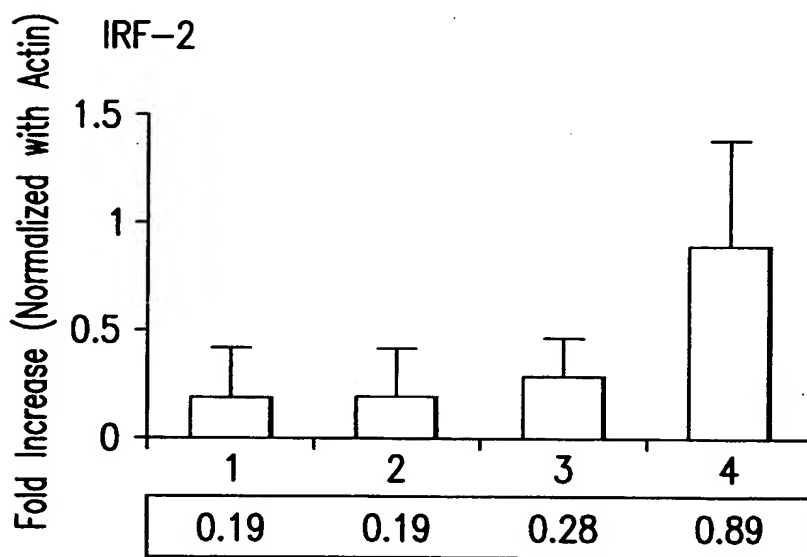
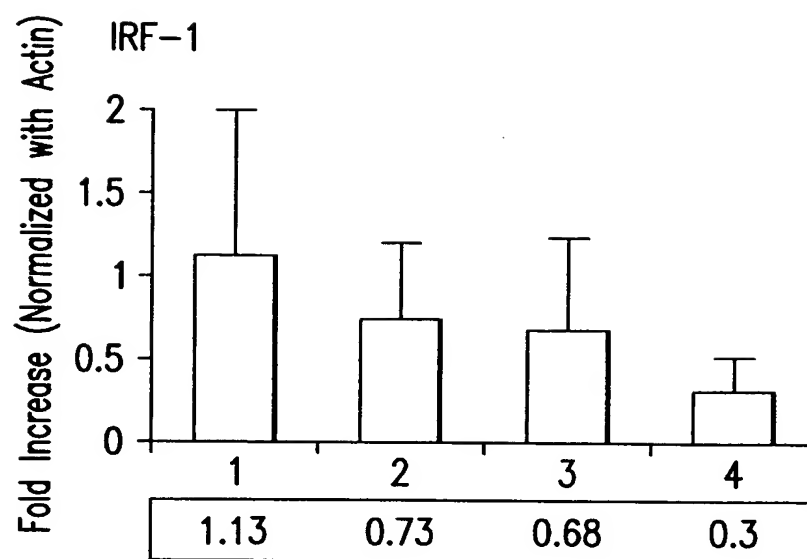


FIG.26

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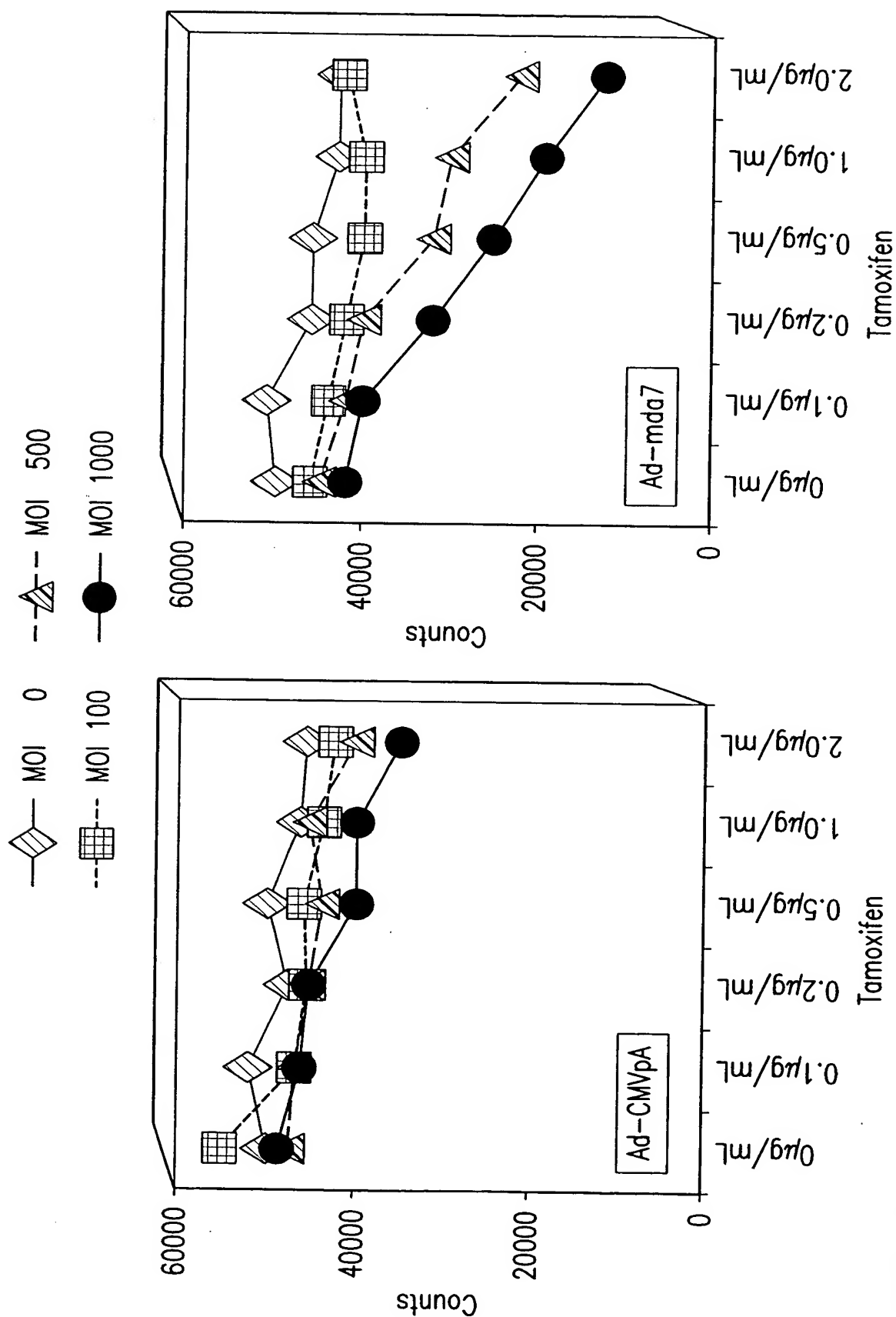


FIG.27

T47D cells

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|             | <u>IL-6</u> | <u>IFN-g</u> | <u>IL-10</u> | <u>GM-CSF</u> | <u>s.Fas</u> | <u>IL-1b</u> | <u>TNF-a</u> | <u>pSTAT3</u> |
|-------------|-------------|--------------|--------------|---------------|--------------|--------------|--------------|---------------|
| Ad-<br>mda7 | ++          | ++           | decrease     | +/-           | ++           | -            | -            | ++            |
| MDA-7       | +           | +++          | decrease     | n.d.          | n.d.         | n.d.         | n.d.         | ++            |

+>+++ : increased cytokine secretion compared to Ad-luc control  
 -: no change; n.d.: not done.  
 Cytokine levels were evaluated using ELISA from cultured cell supernatants taken 24-72 hr post treatment.  
 pSTAT3 results were analyzed 4hr after addition of MDA-7 protein and 24 hr after Ad-mdm7 treatment using IHC.

FIG.28

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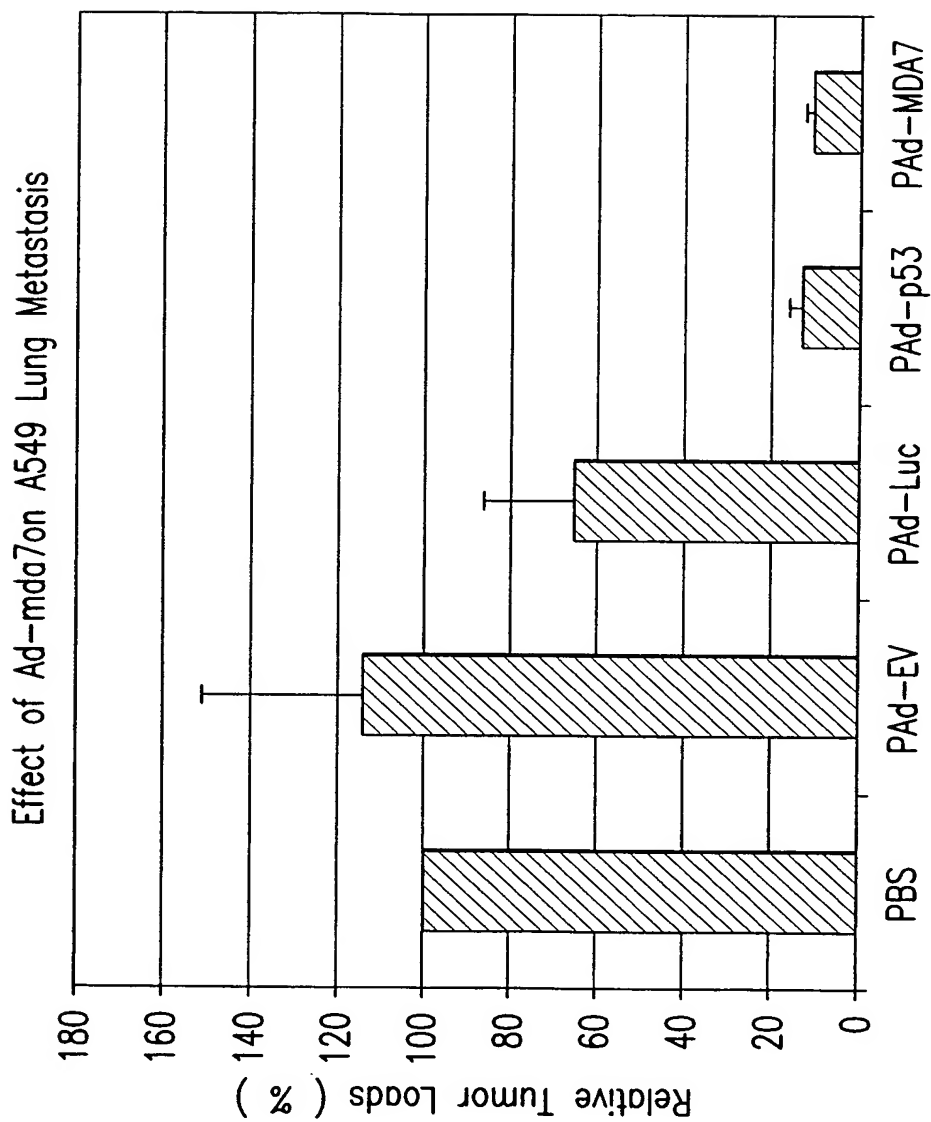


FIG.29

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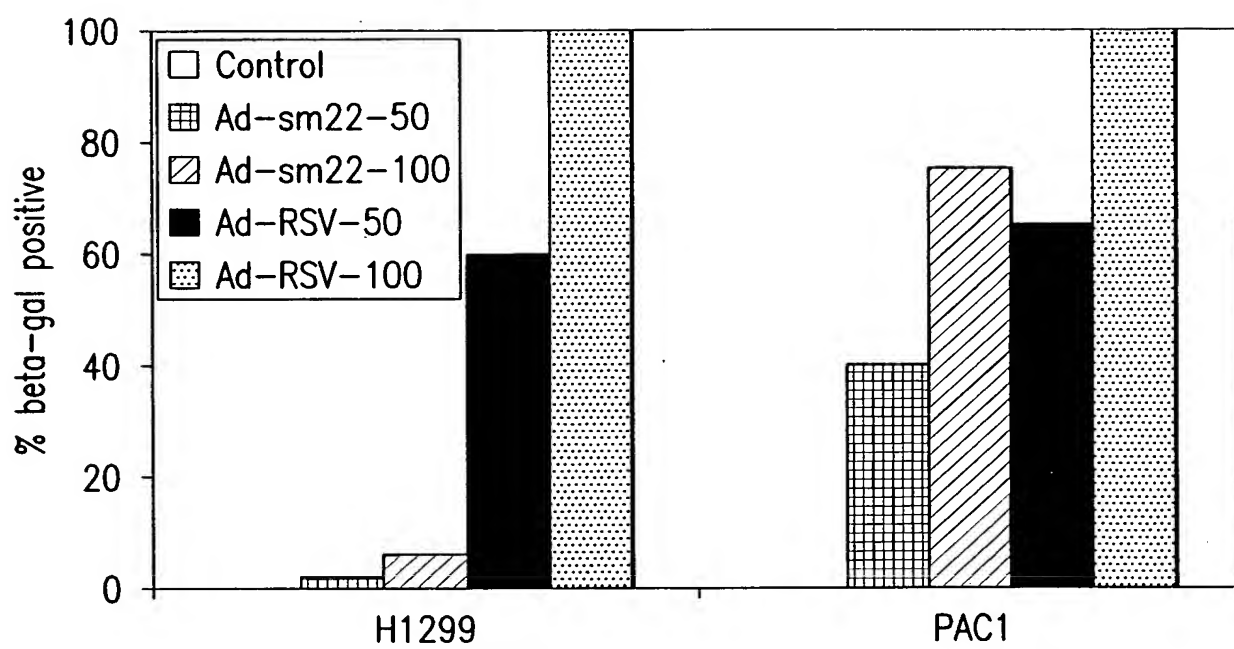


FIG.30

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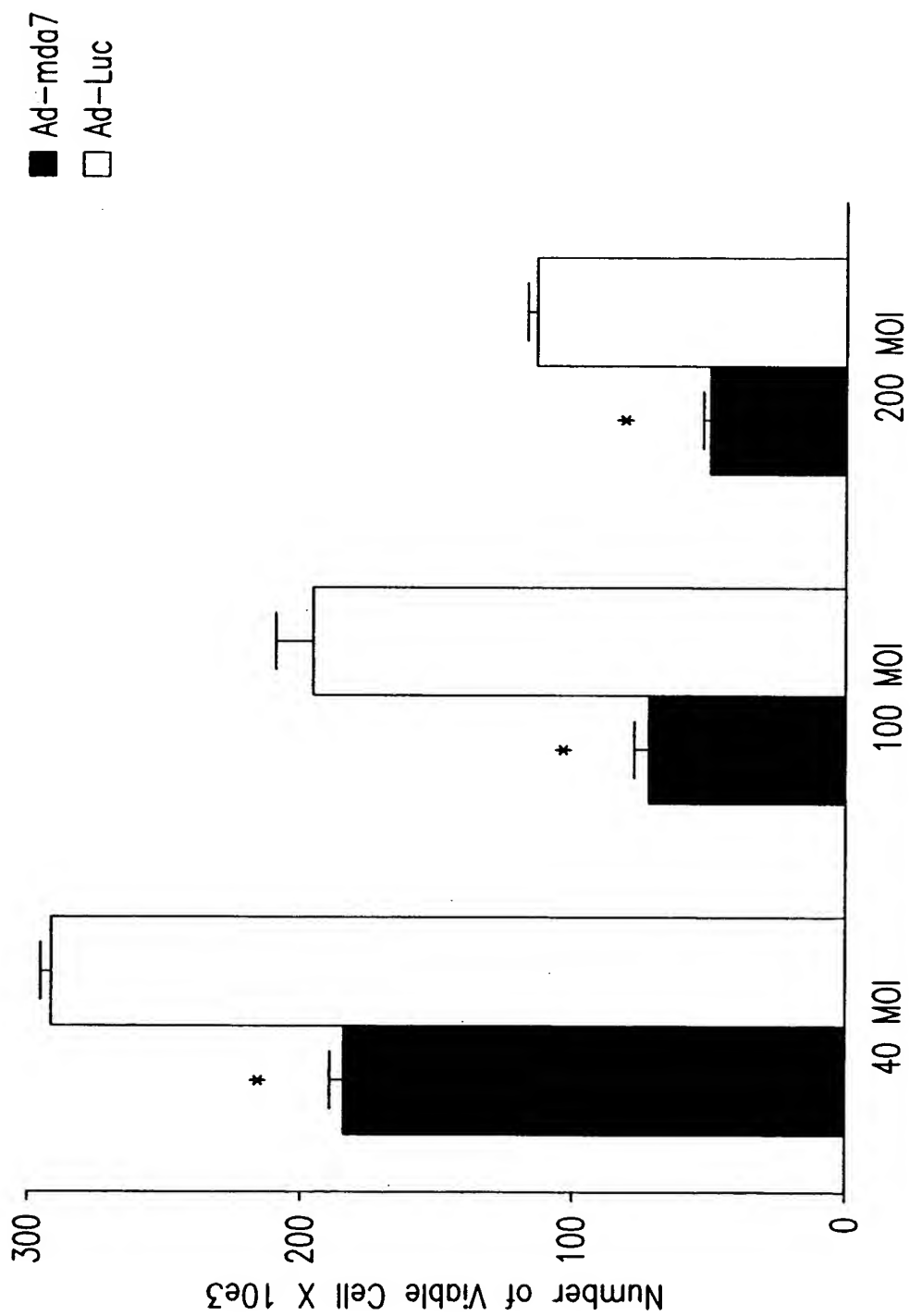


FIG.31

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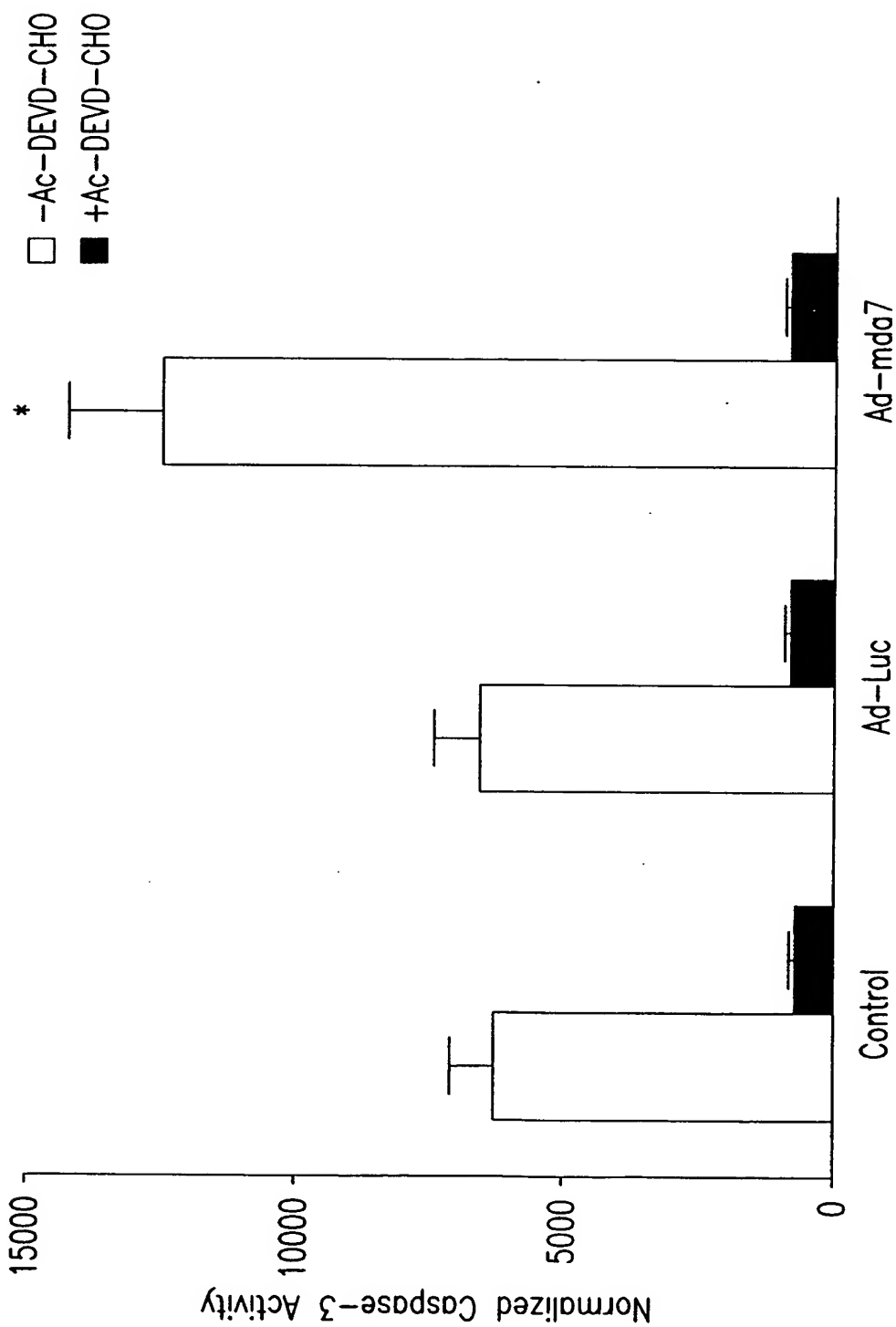


FIG.32A

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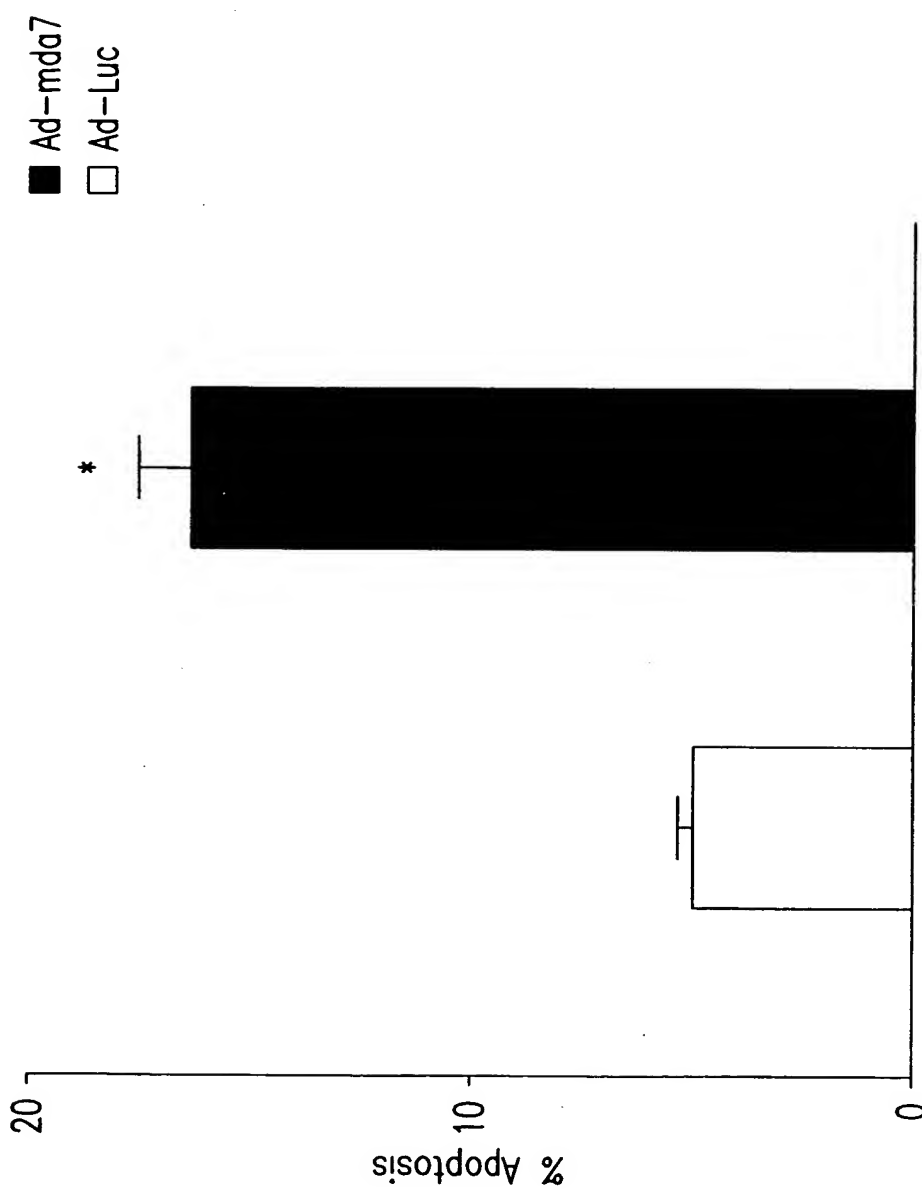


FIG.32B



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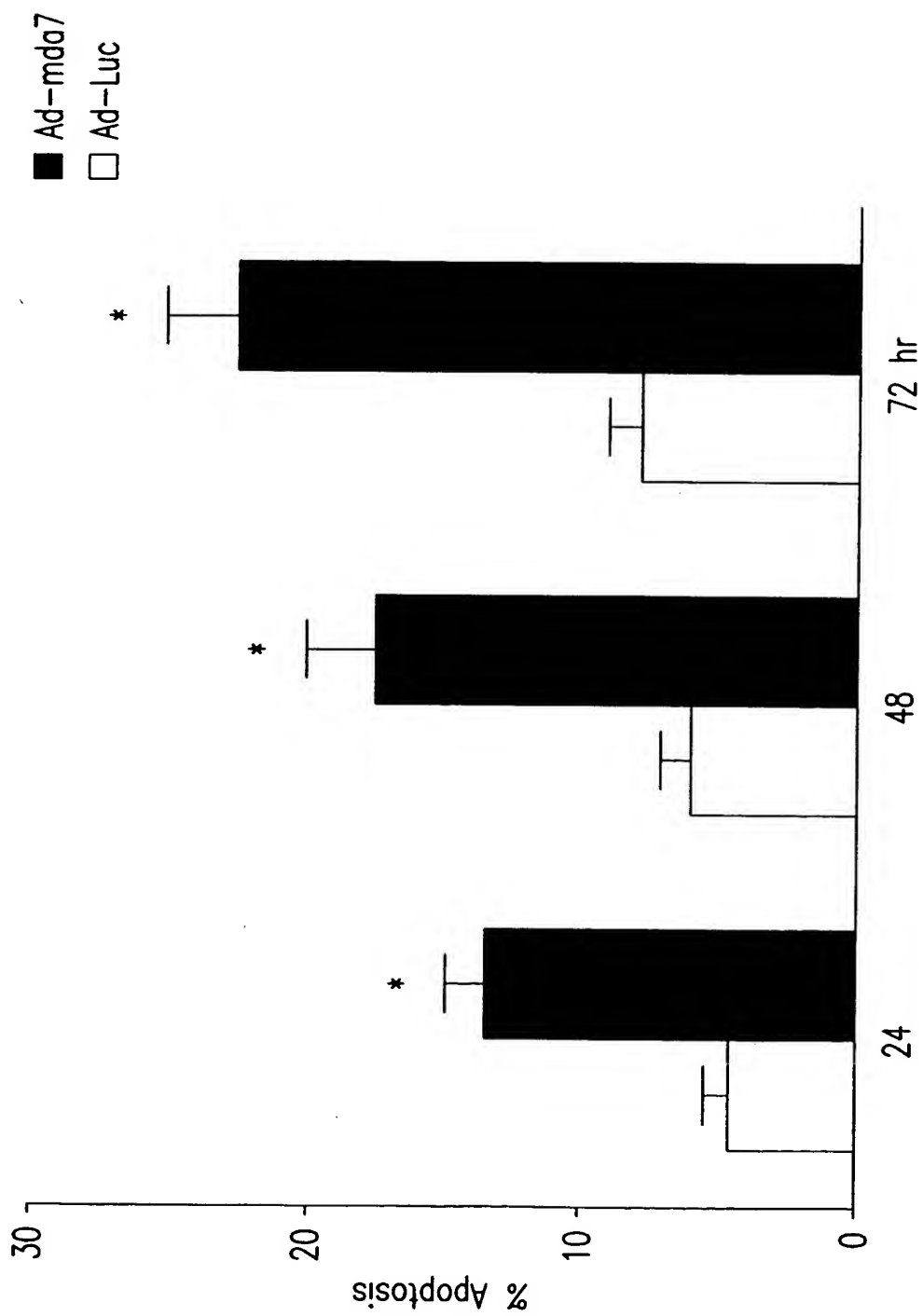


FIG.32C

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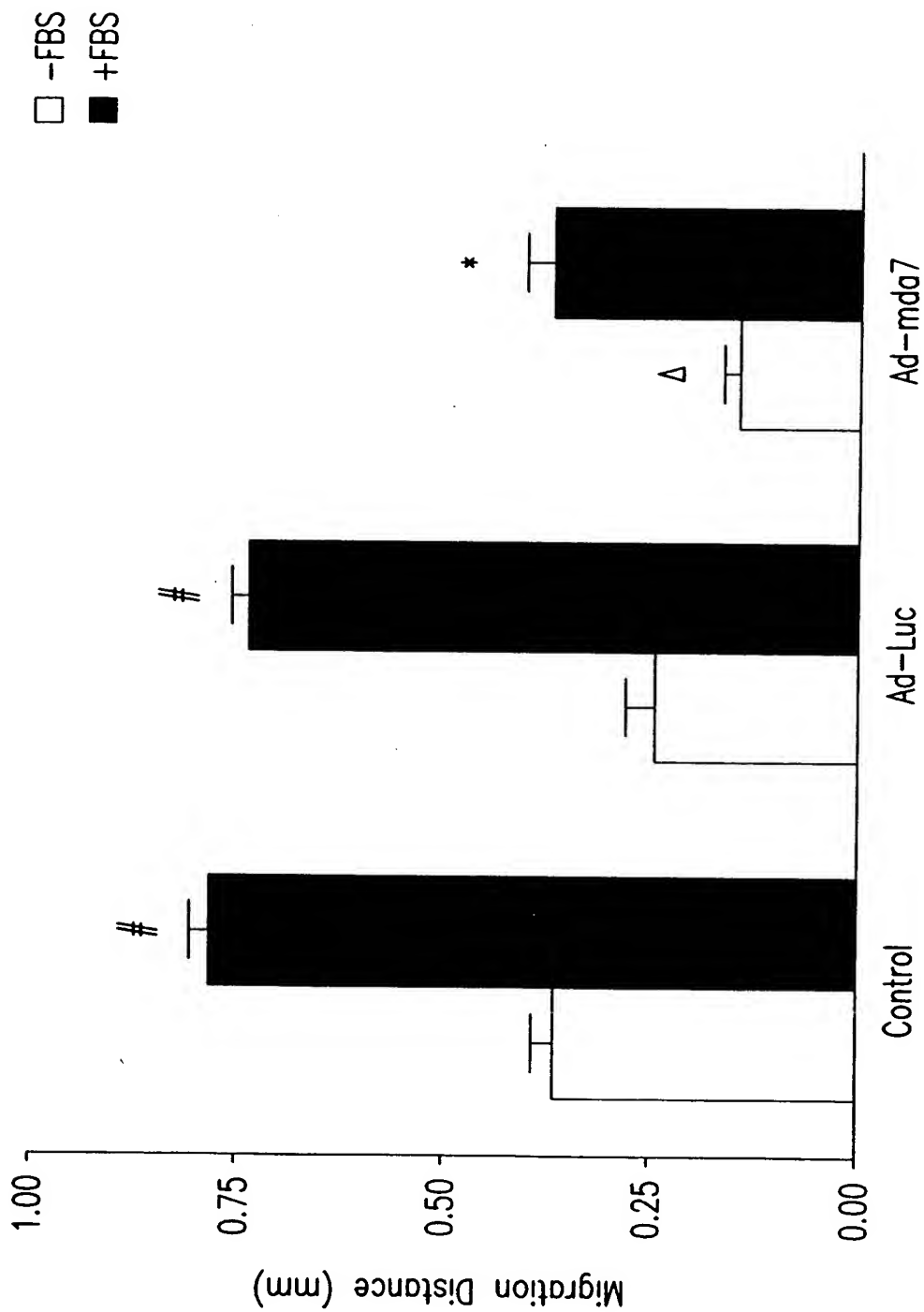


FIG.33

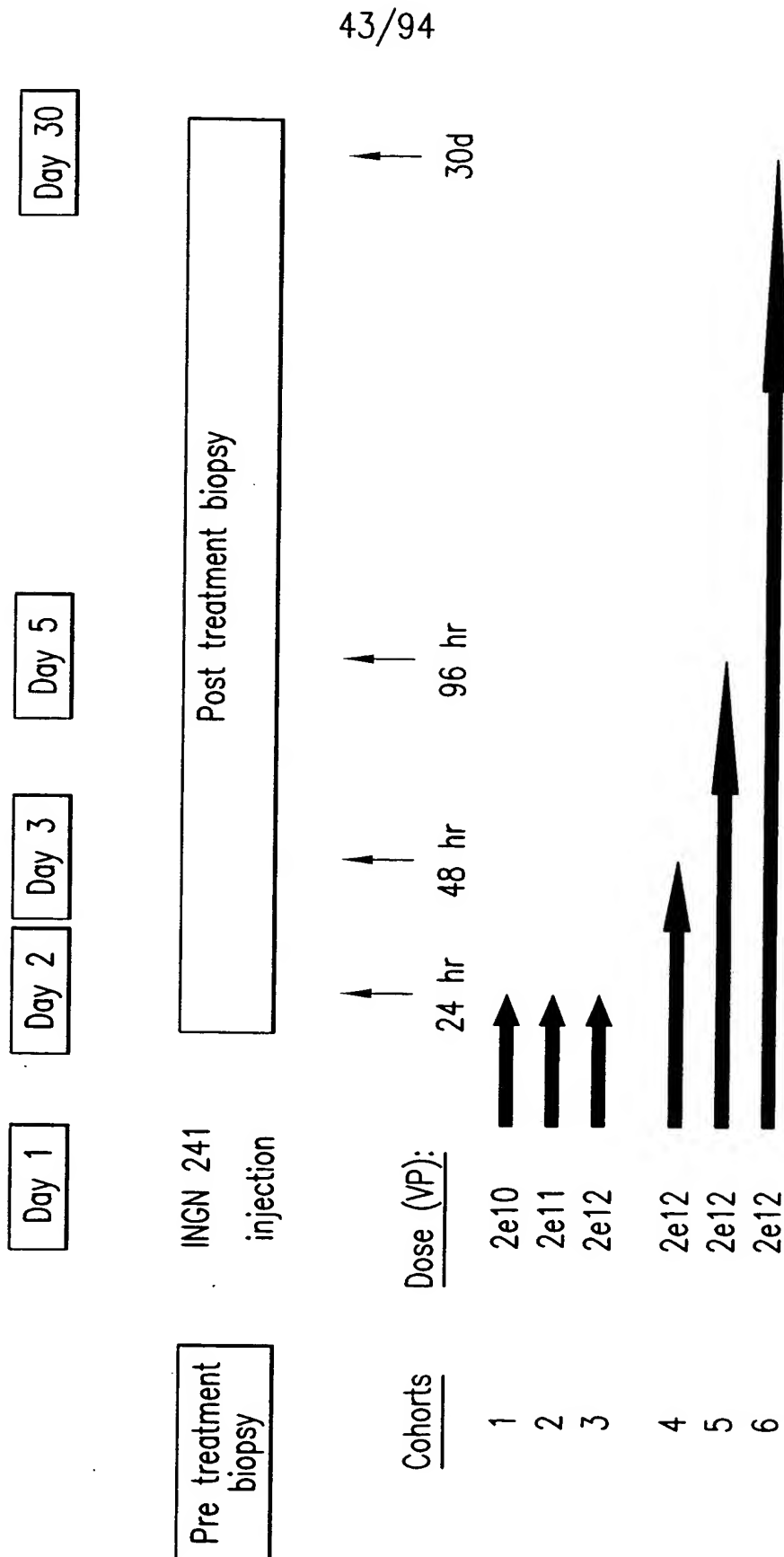
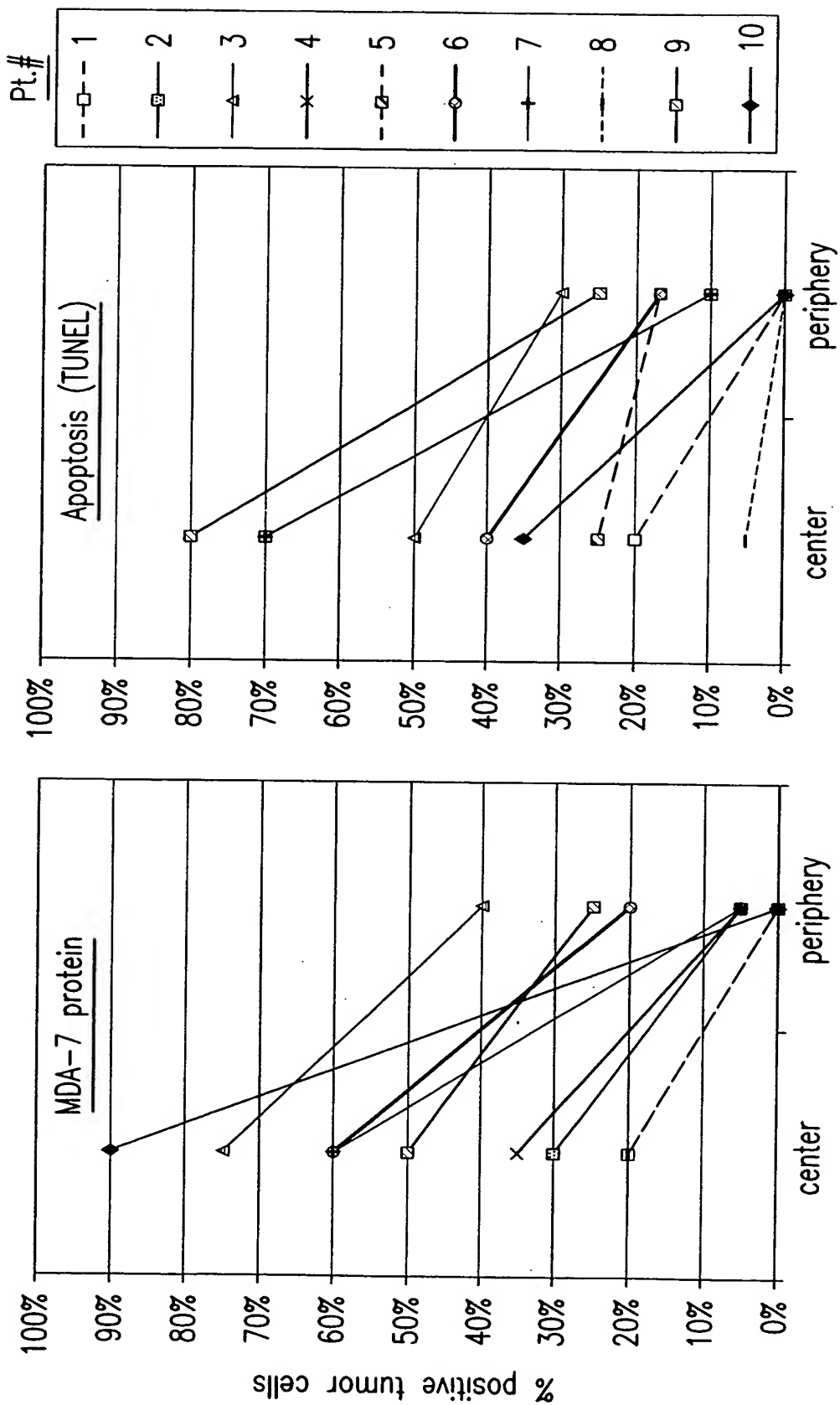


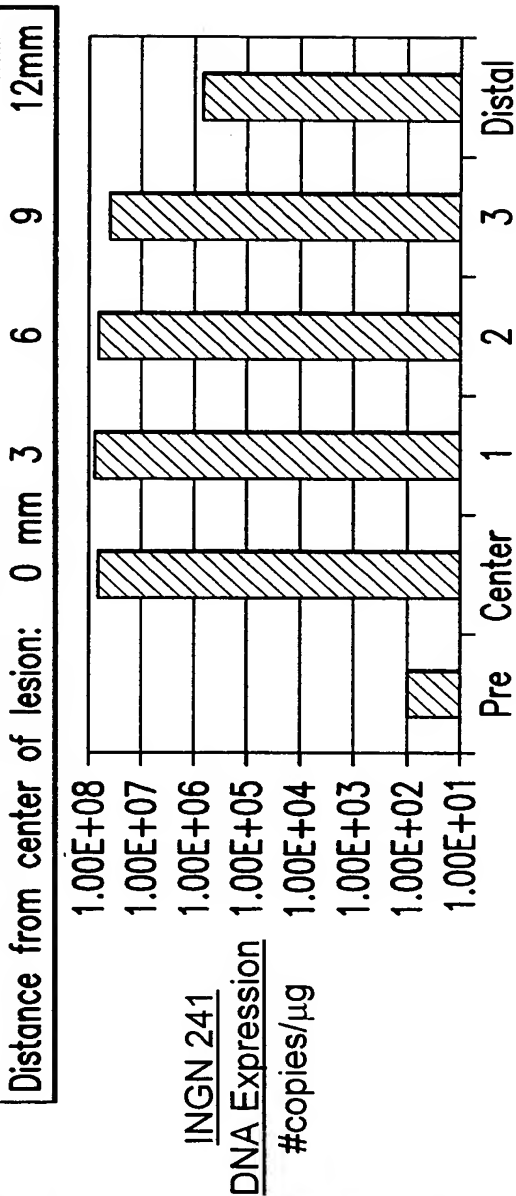
FIG.34

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MDA-7/ TUNEL correlation  $p < 0.01$

FIG.35



MDA-7 protein expression

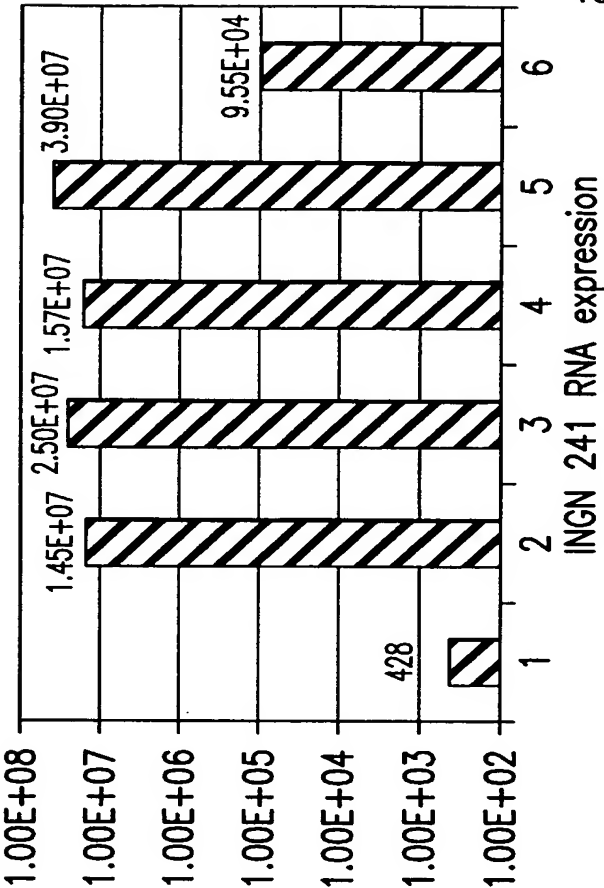
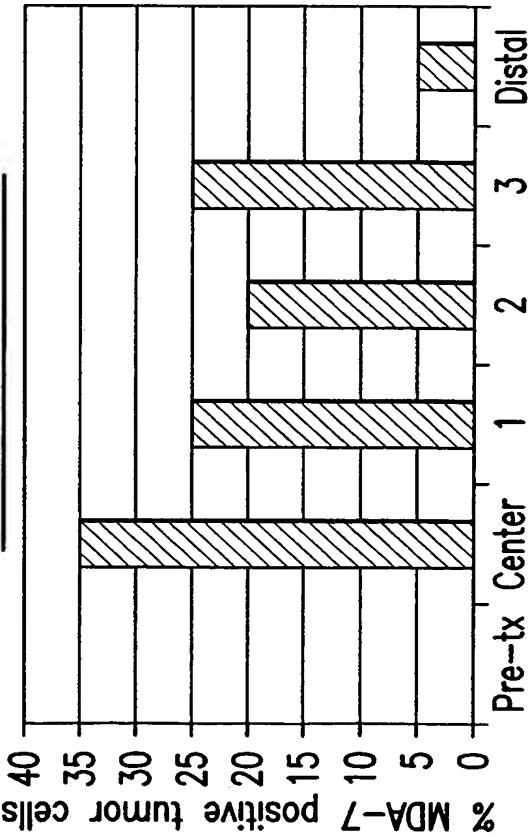


FIG.36

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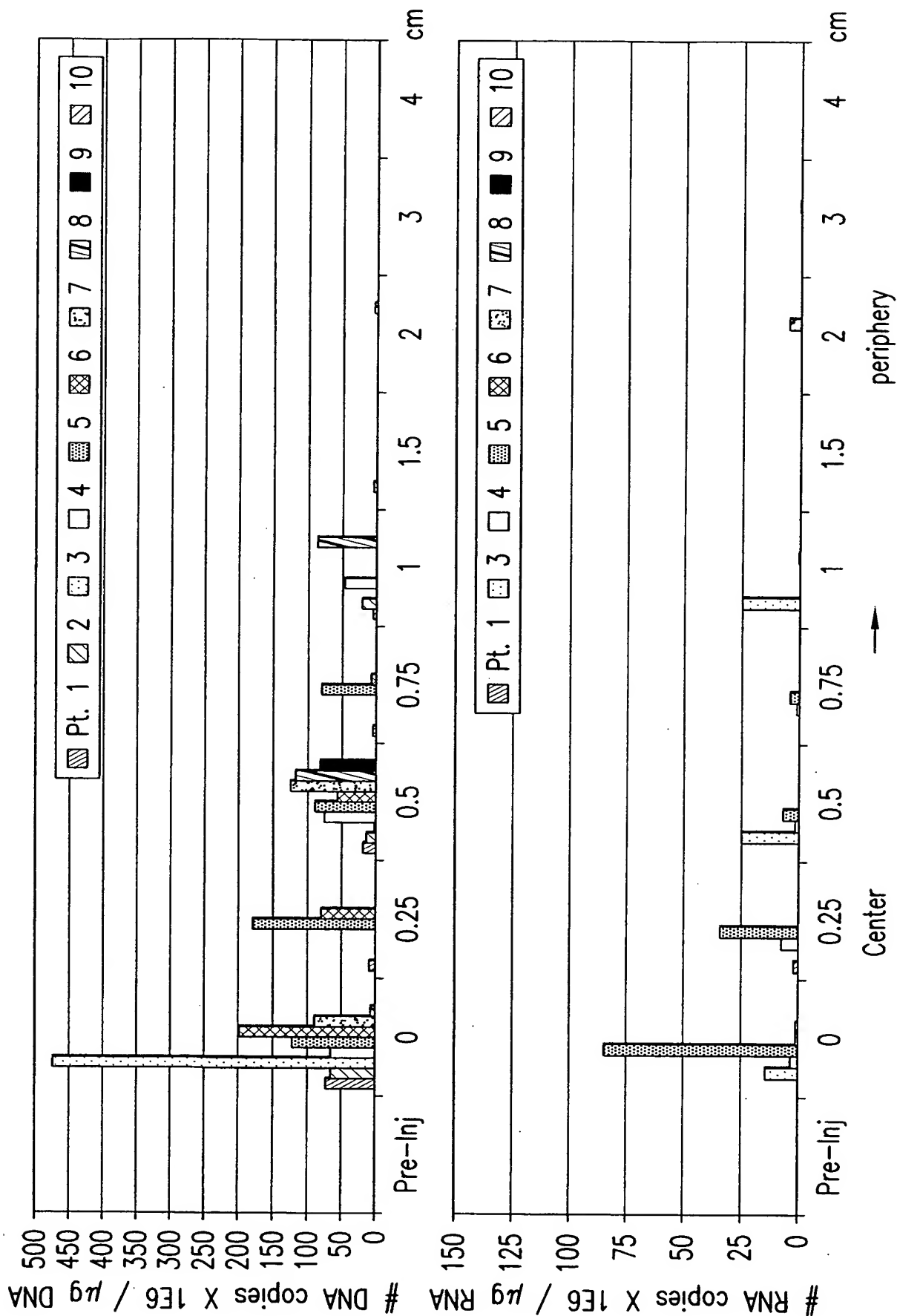


FIG.37

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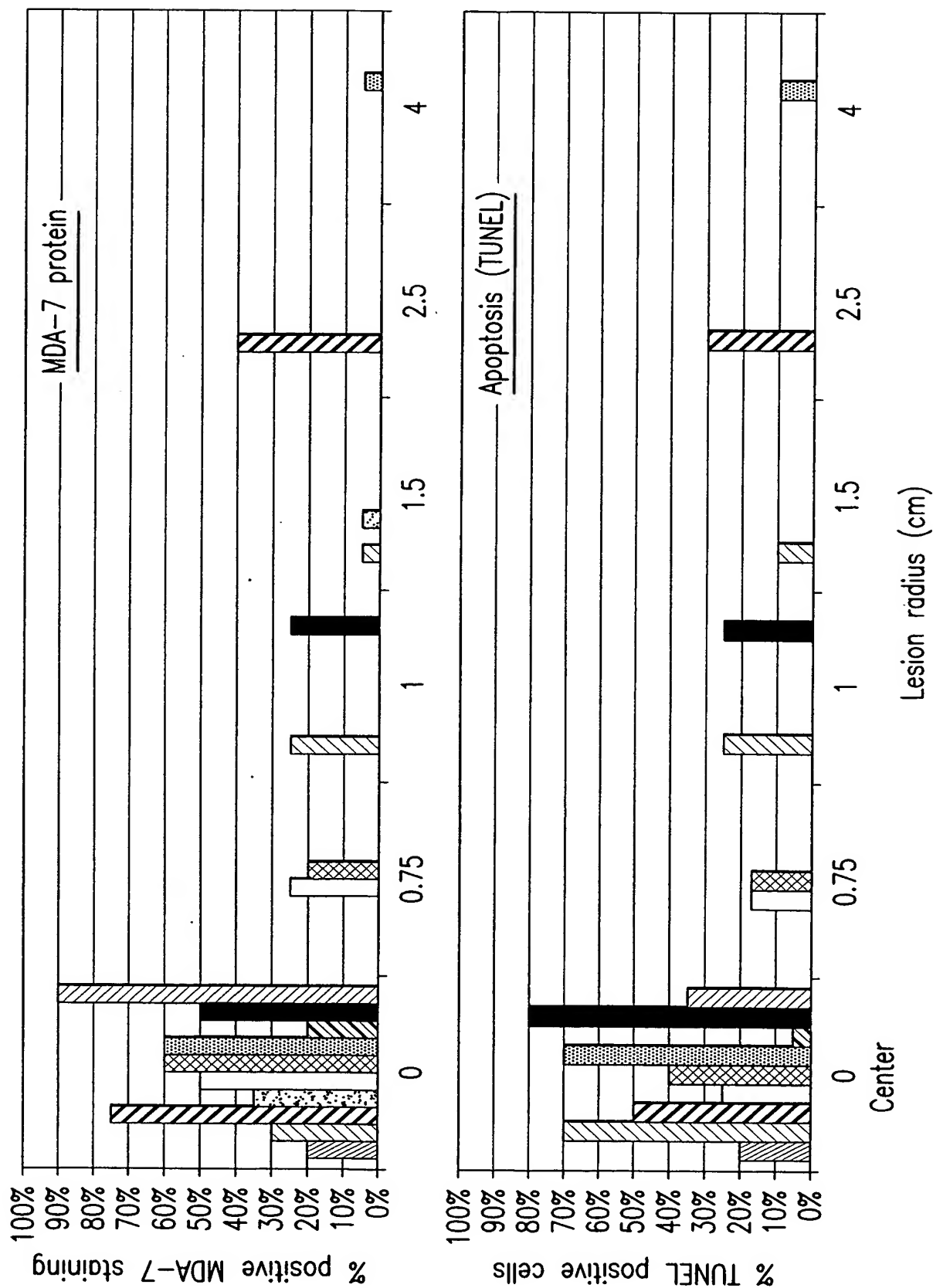


FIG.38

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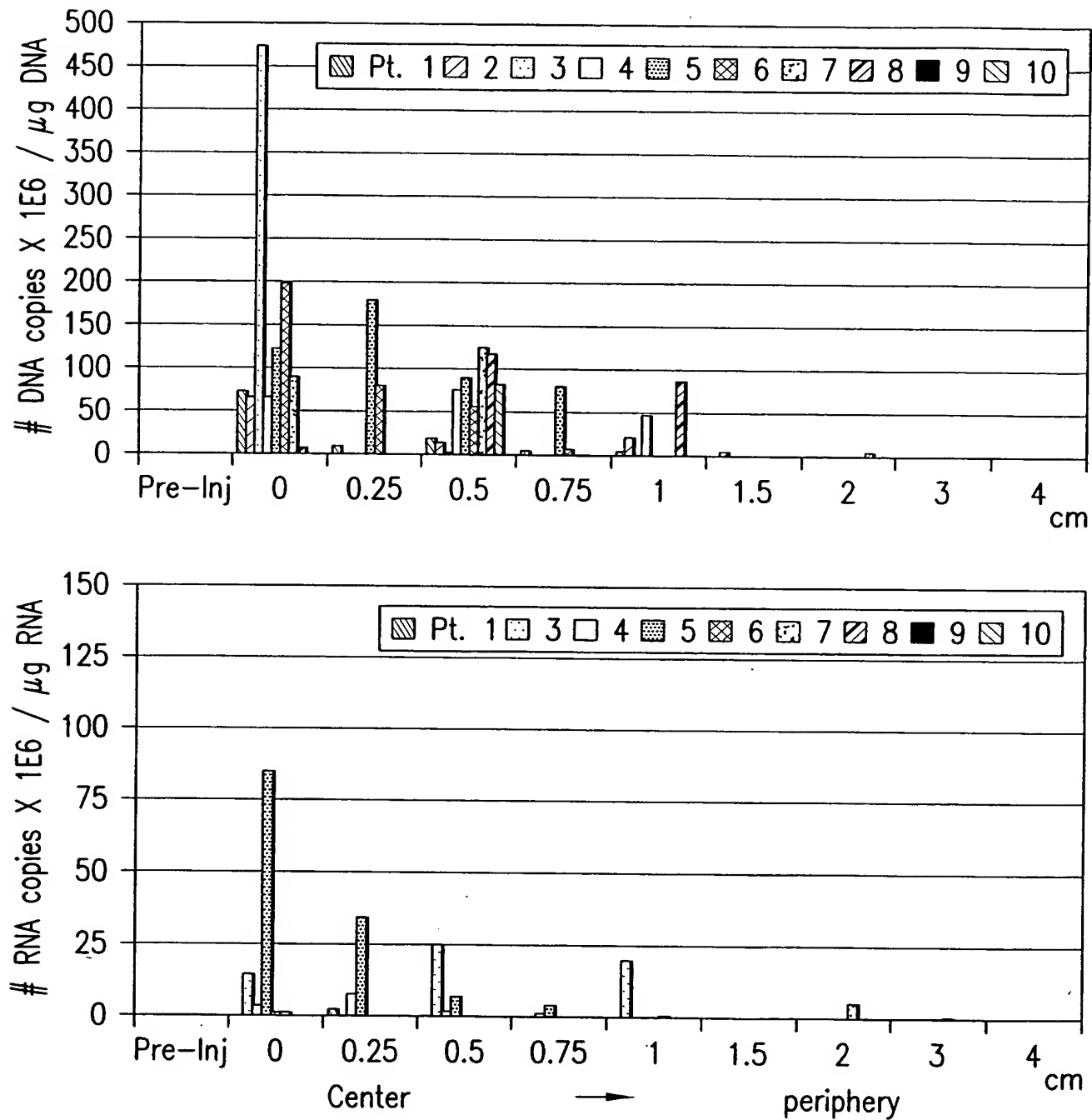


FIG.39A



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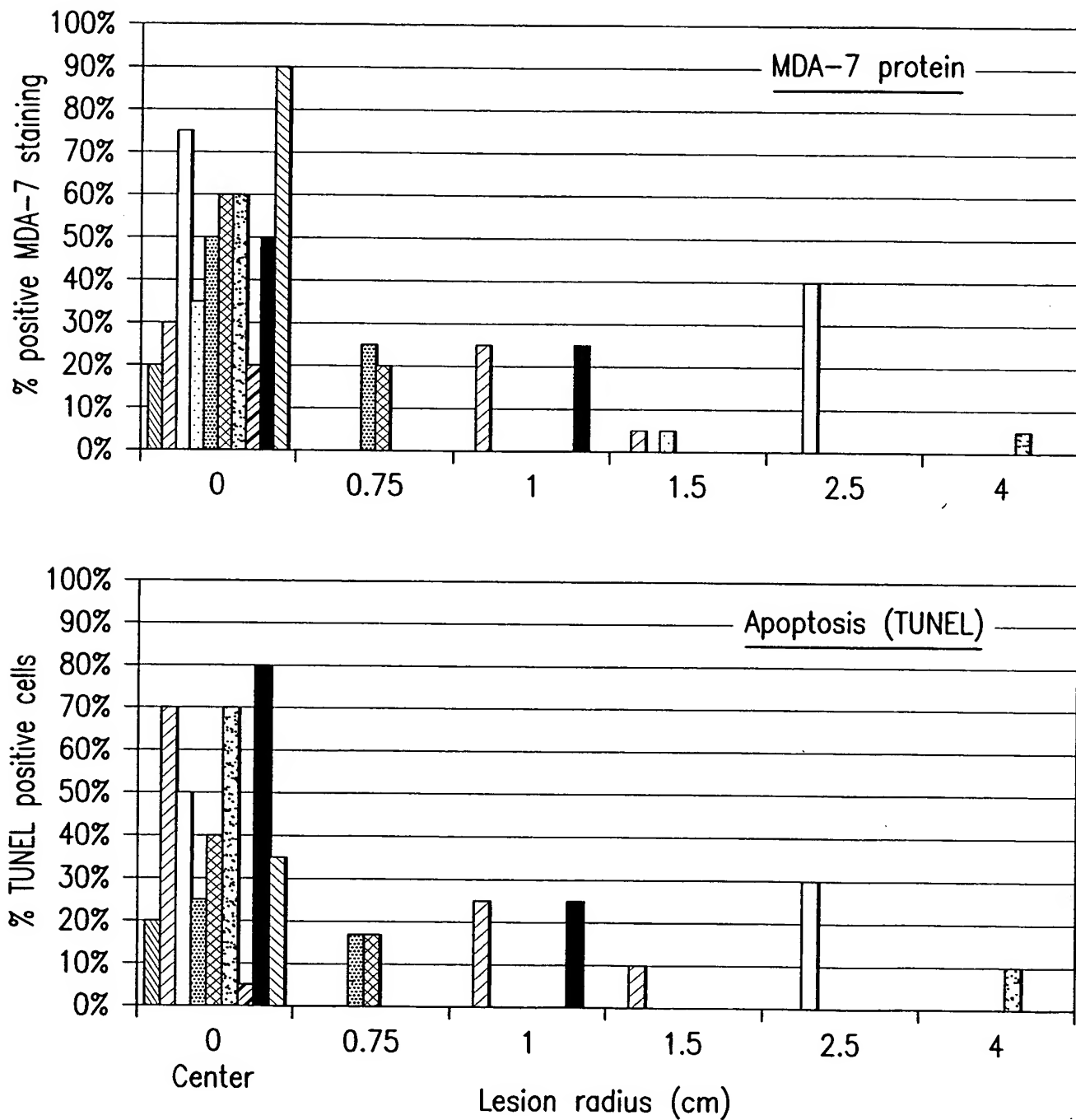


FIG.39B

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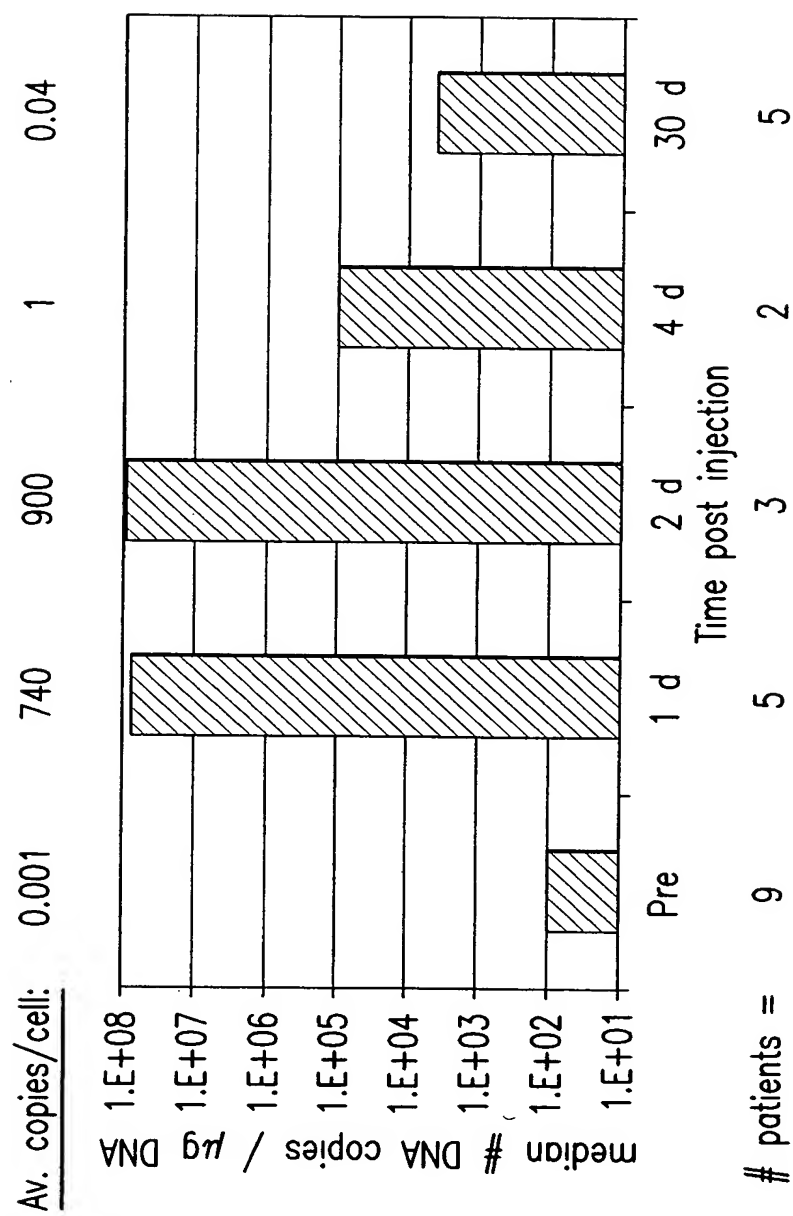


FIG.40

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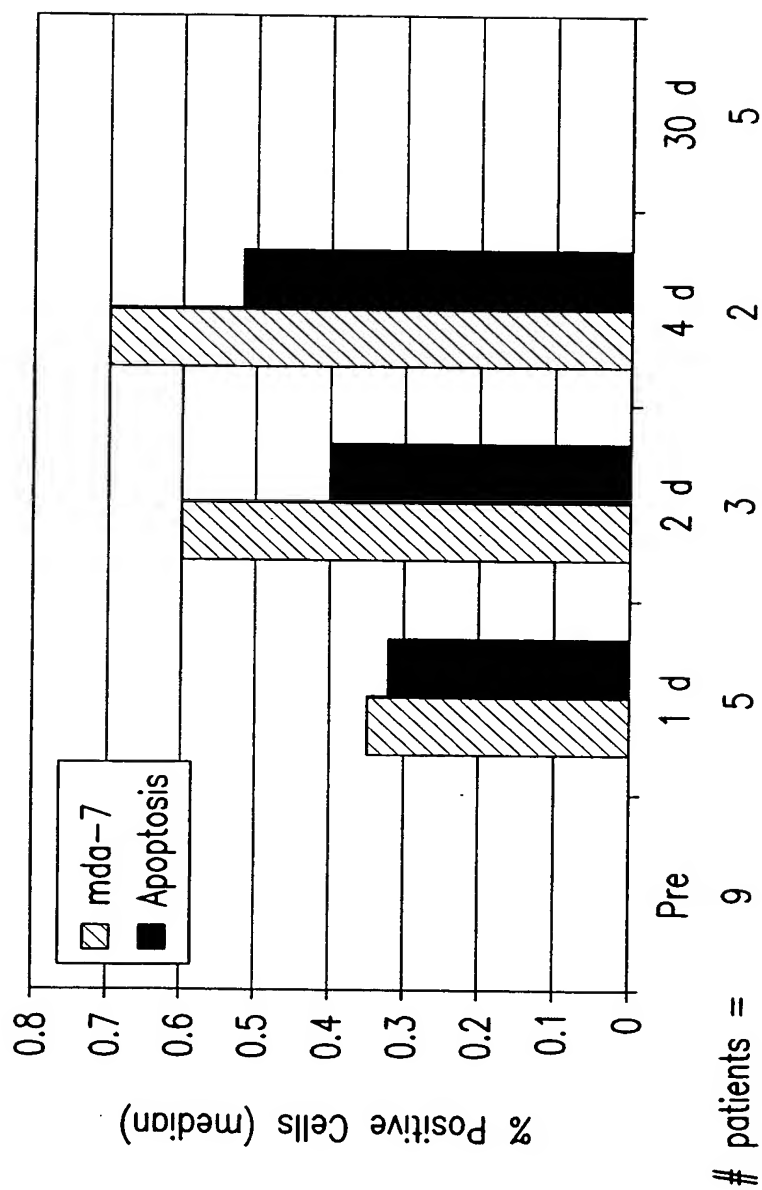


FIG.41

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| Single Dose (2E12 vp)             |                   | Clinical Activity (Y/N) |       |         |                          |
|-----------------------------------|-------------------|-------------------------|-------|---------|--------------------------|
| Patient                           | Primary Histology | # injections            | Local | Remote  | Comment                  |
| 71                                | Renal Cell Ca     | 1                       | N     | N       | Withdrew prior to day 30 |
| 72                                | TCC Bladder       | 1                       | N     | N       |                          |
| 73                                | Melanoma          | 1                       | N     | N       |                          |
| 74                                | Colo--Rectal      | 1                       | N     | N       | Withdrew prior to day 30 |
| 75                                | SCCHN             | 1                       | N     | N       |                          |
| 76                                | SCCHN             | 1                       | N     | N       |                          |
| 77                                | SCCHN             | 1                       | N     | N       |                          |
| Repeat Dose (2E12 vp biweekly x3) |                   |                         |       |         |                          |
| 81                                | AdenoCa           | 3                       | -     | -       | Withdrew                 |
| 83                                | Melanoma          | 12                      | Y*    | Y**     | CR                       |
| 84                                | Melanoma          | 12                      | N     | N       | Stable Disease           |
| 85                                | SCCHN             | 6                       | Y     | N       | Central necrosis         |
| 86                                | NSCLC             | 3                       | -     | -       | Not eval                 |
| 87                                | SCCHN             | 6                       | Y     | pending | On study                 |
| 88                                | Melanoma          | 6                       | Y     | pending | On study                 |

\* Three lesions treated consecutively: pCR in 1st, clinCR in 2nd, regression in 3rd  
 \*\*marked crythema around remote cutaneous lesion

FIG.42

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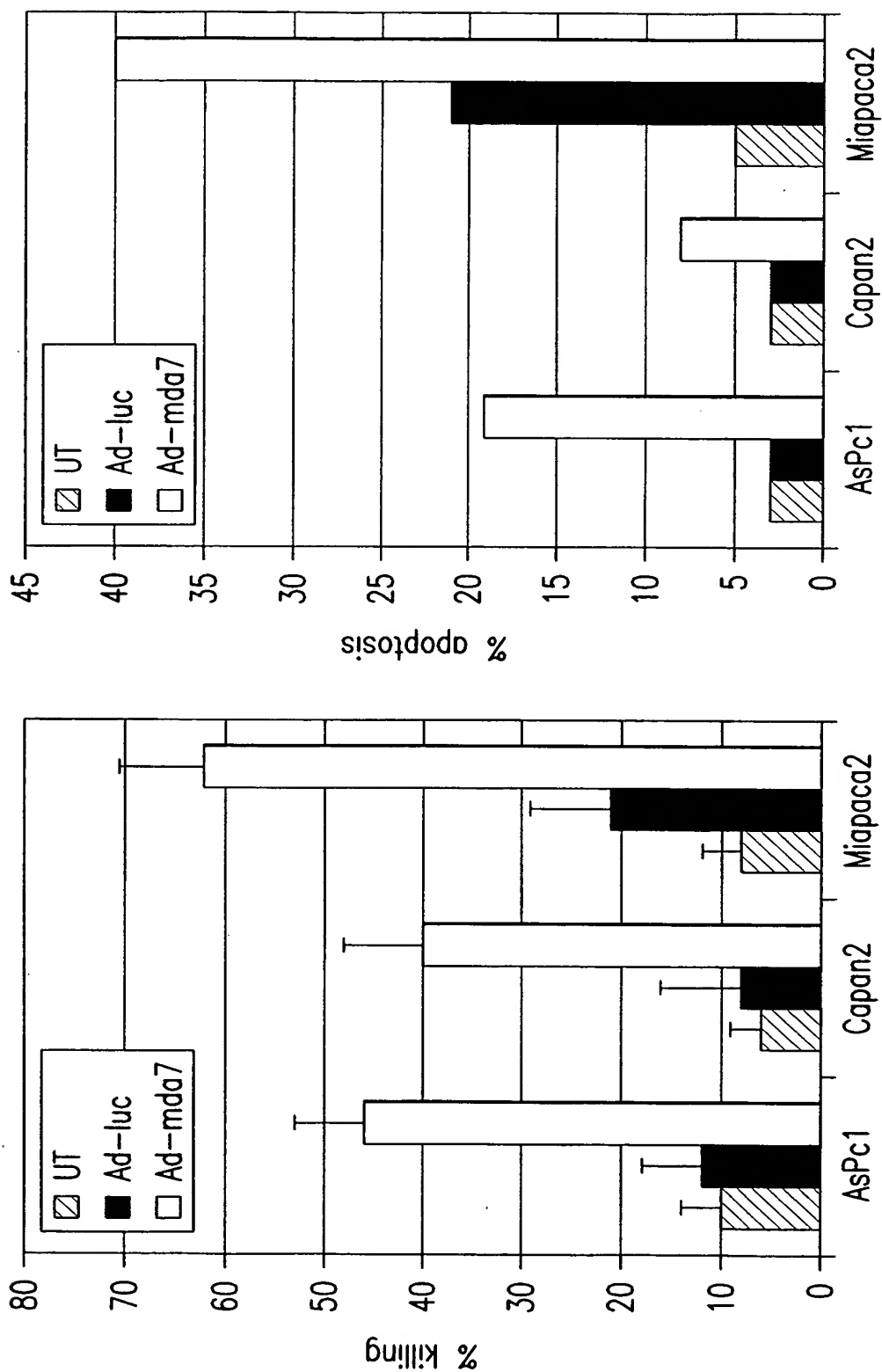


FIG.43

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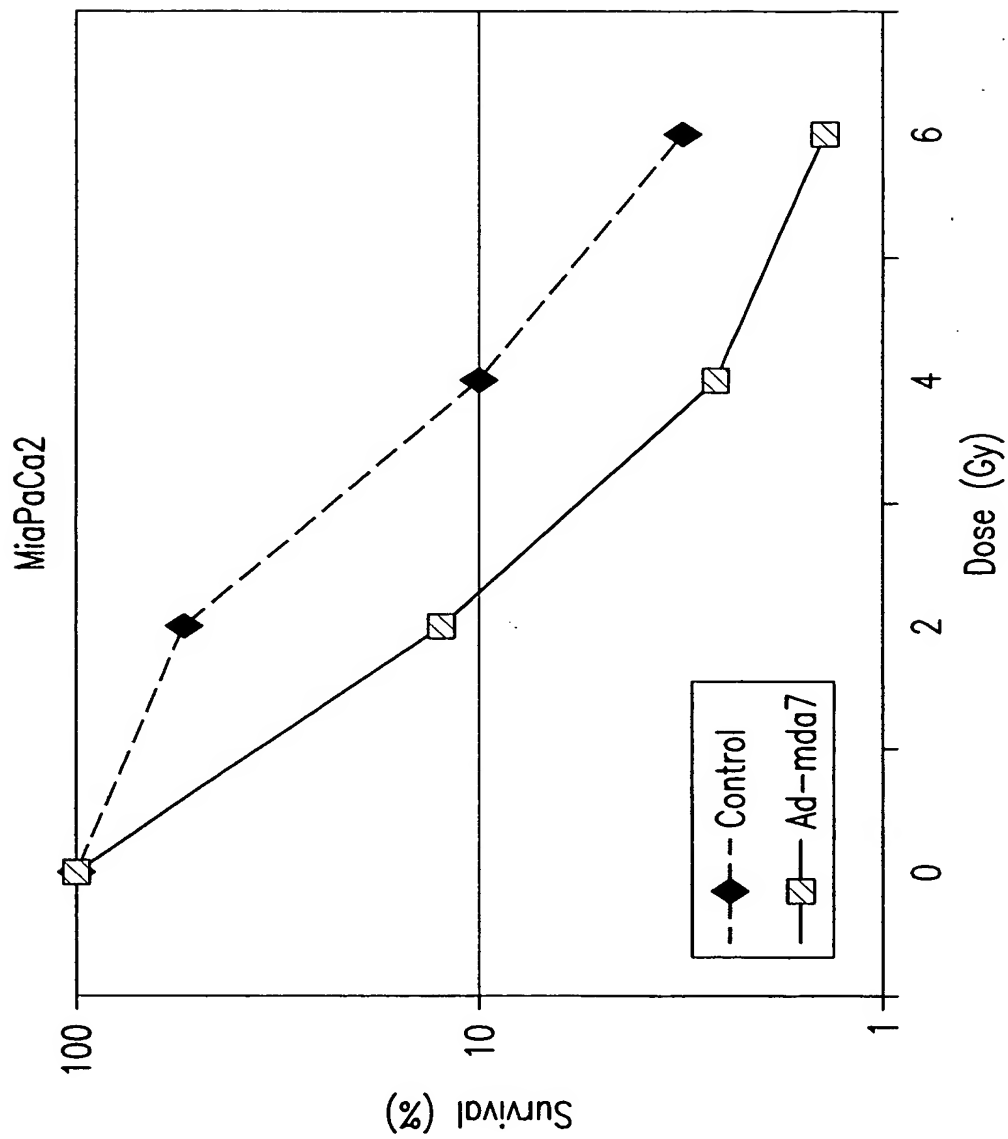


FIG.44

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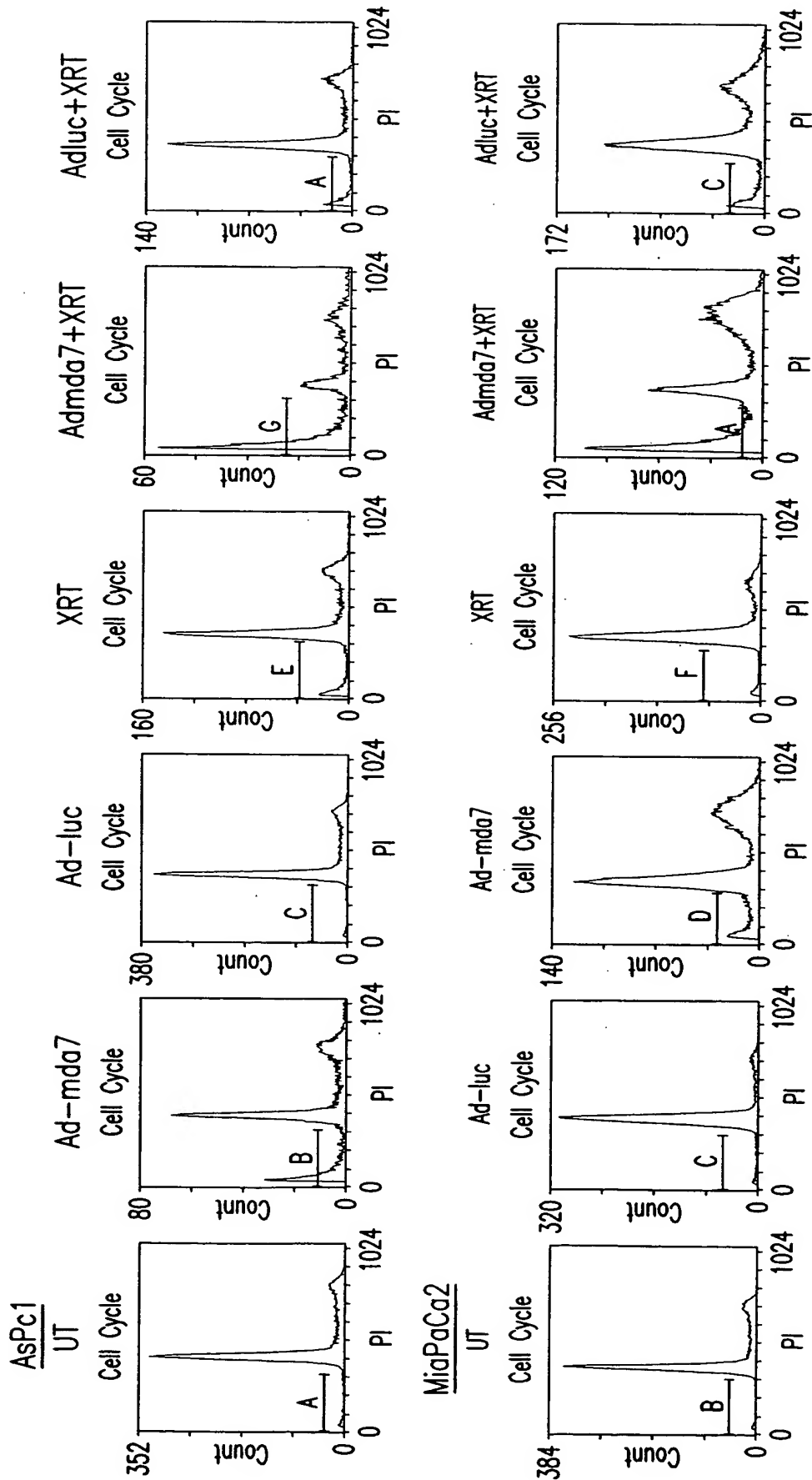


FIG.45

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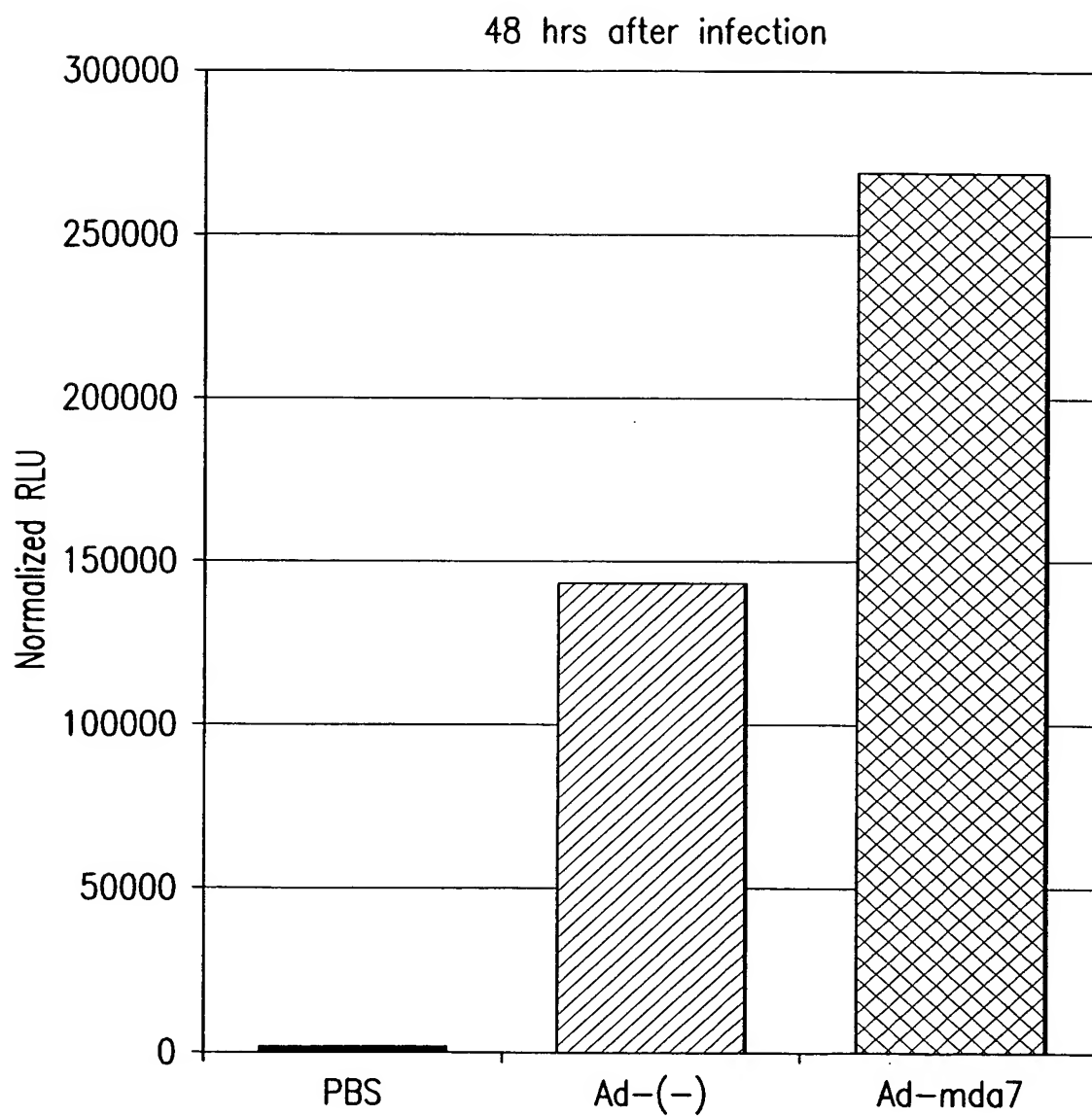


FIG.46



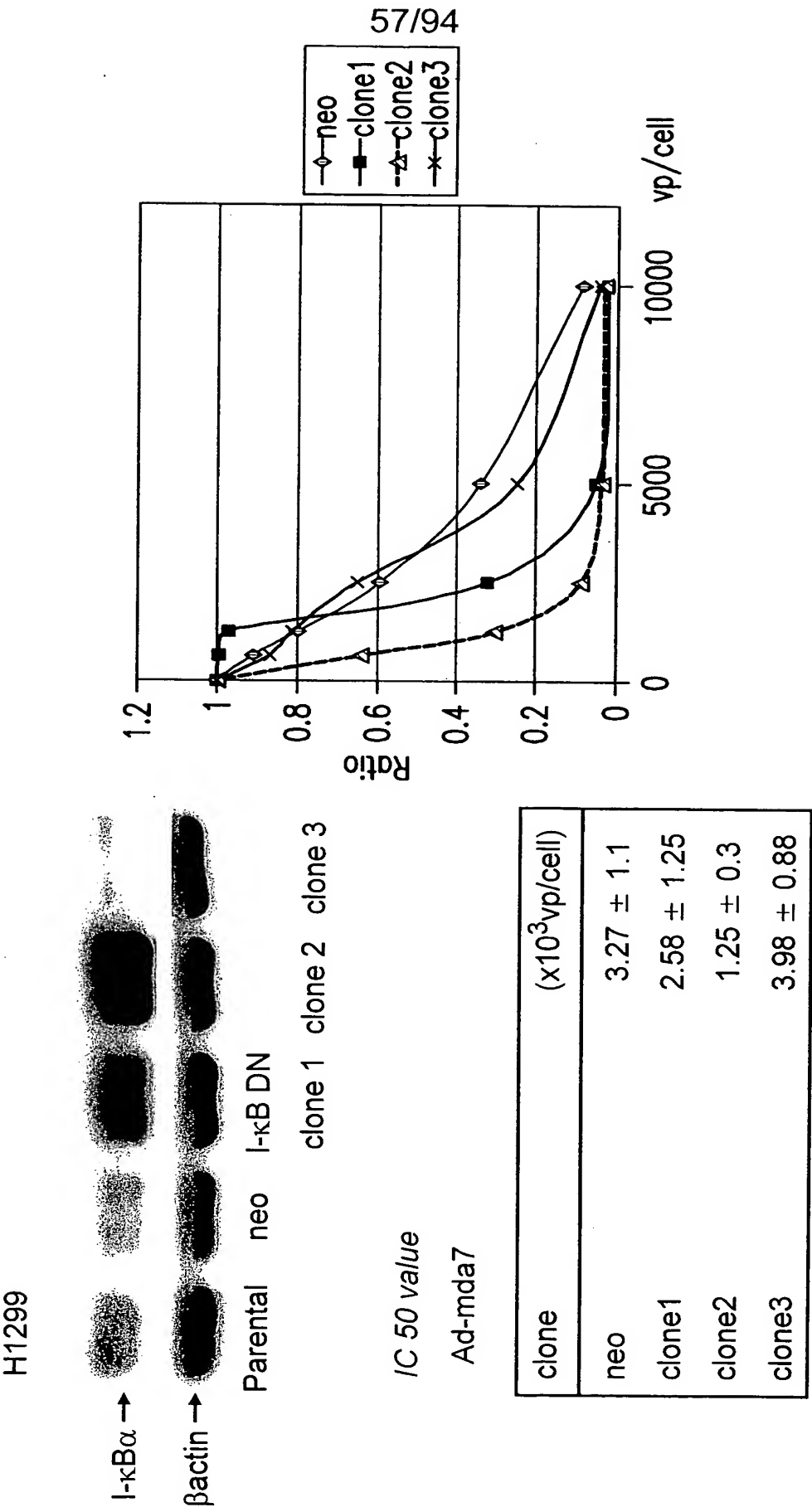


FIG.47

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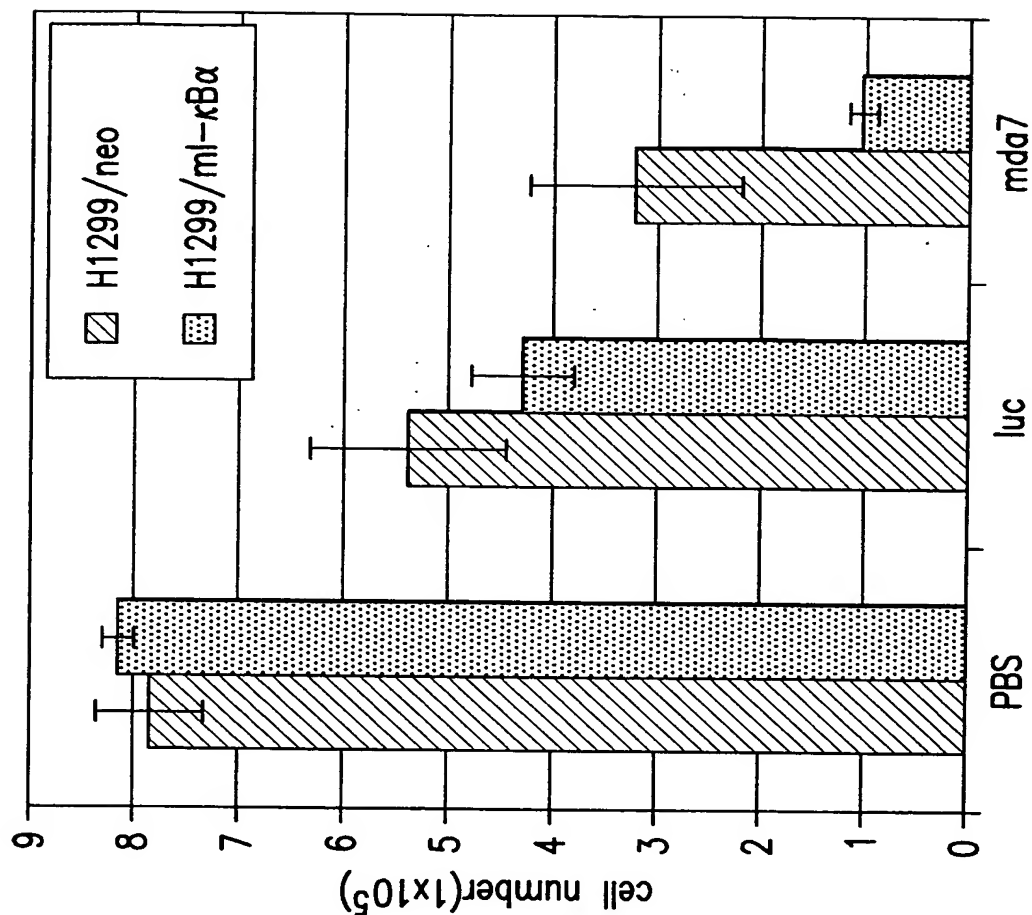
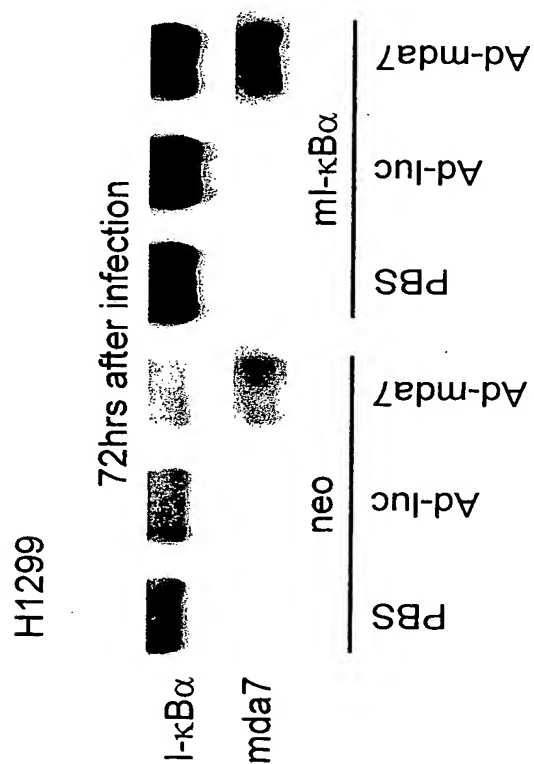


FIG.48



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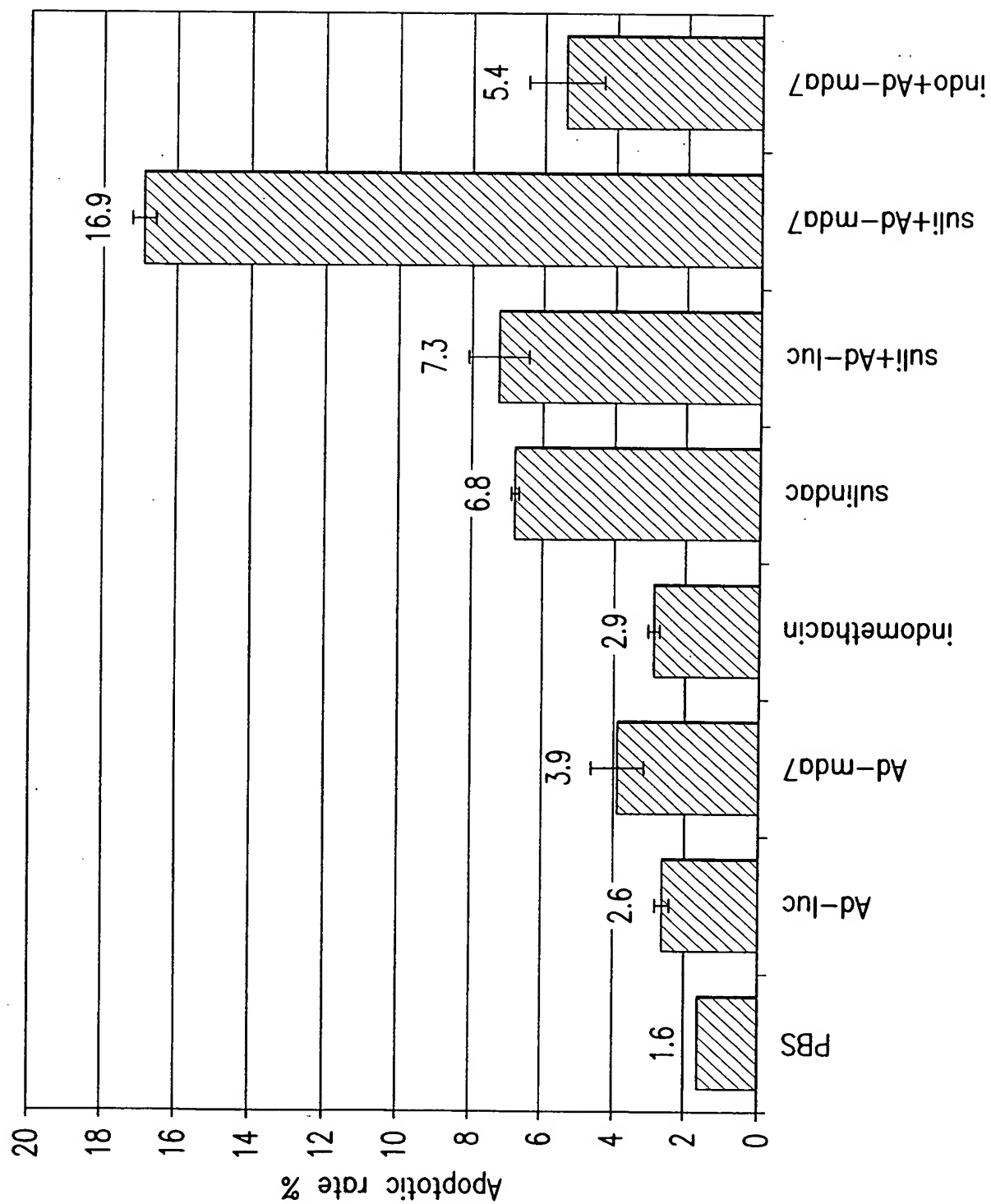


FIG. 49A

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---◆--- PBS  
 ---■--- Ad-luc  
 ---▲--- Ad-mda7

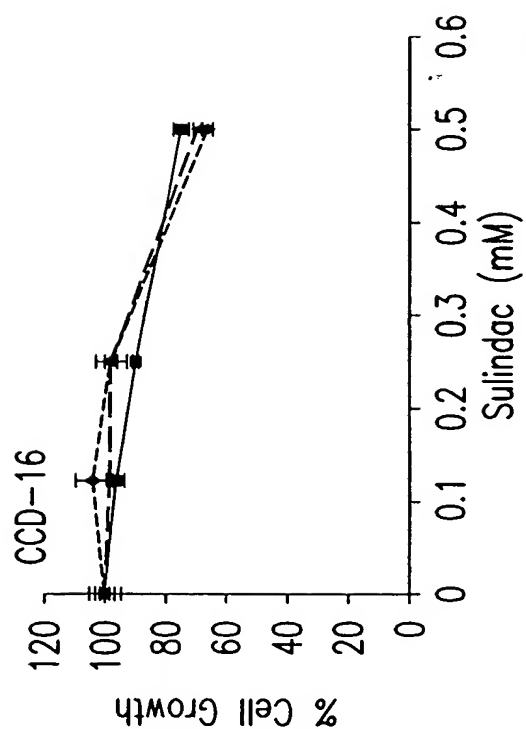
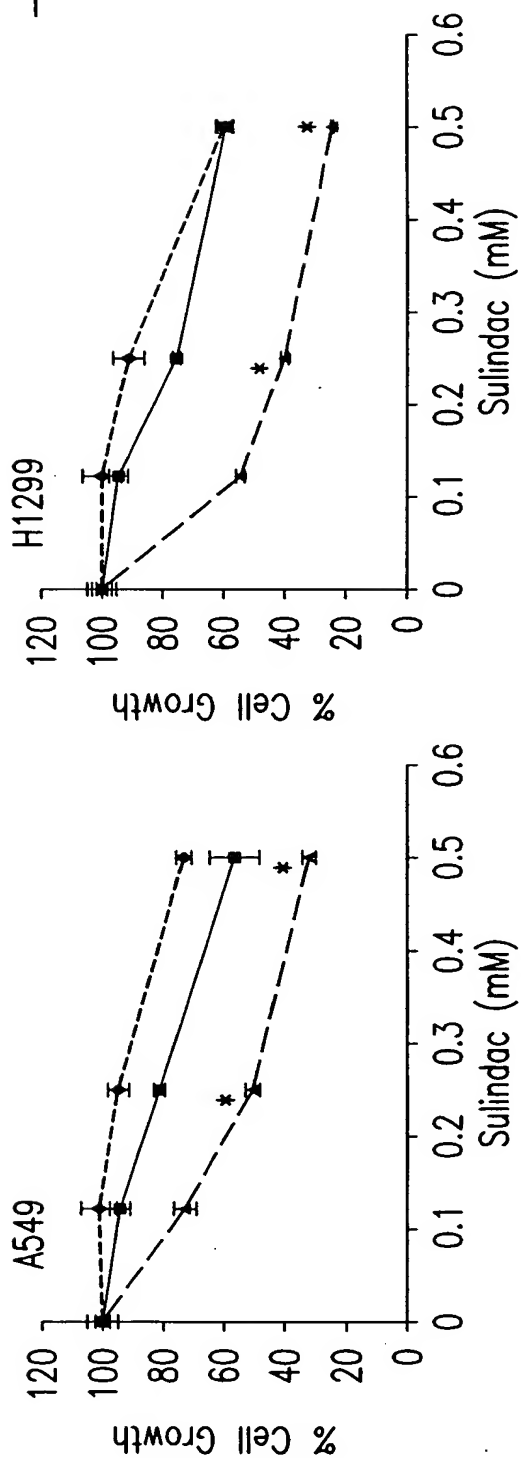


FIG.49B

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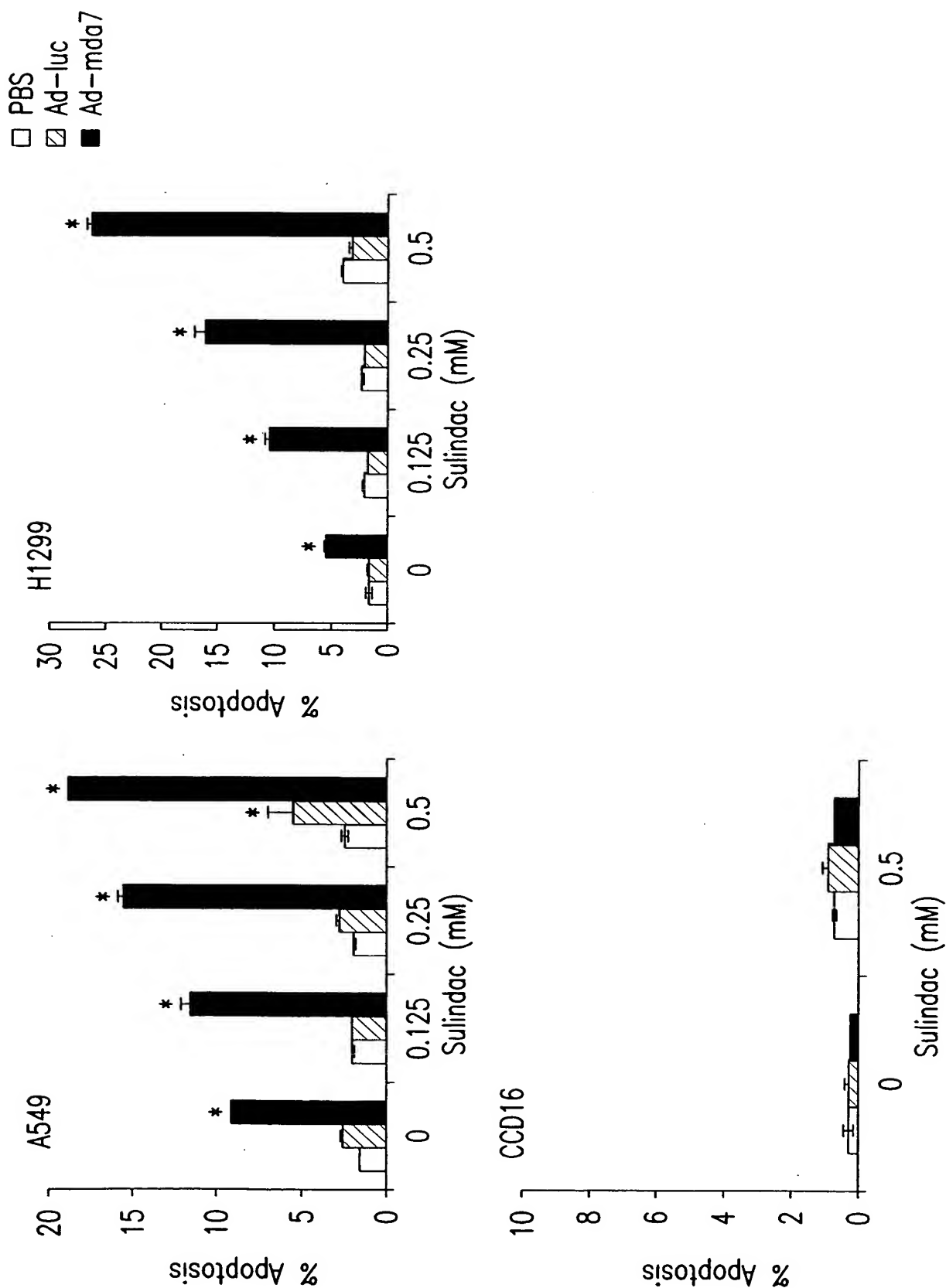


FIG.49C

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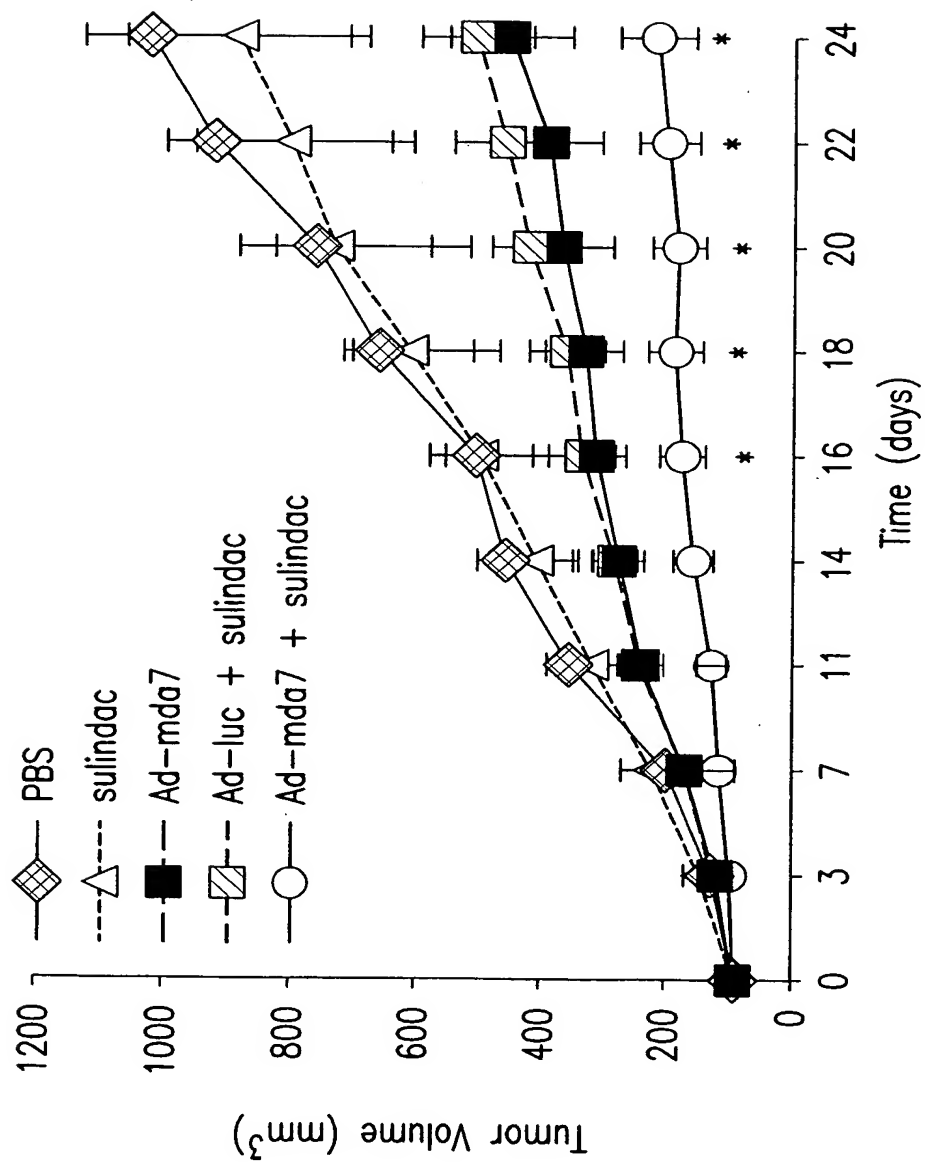


FIG.49D

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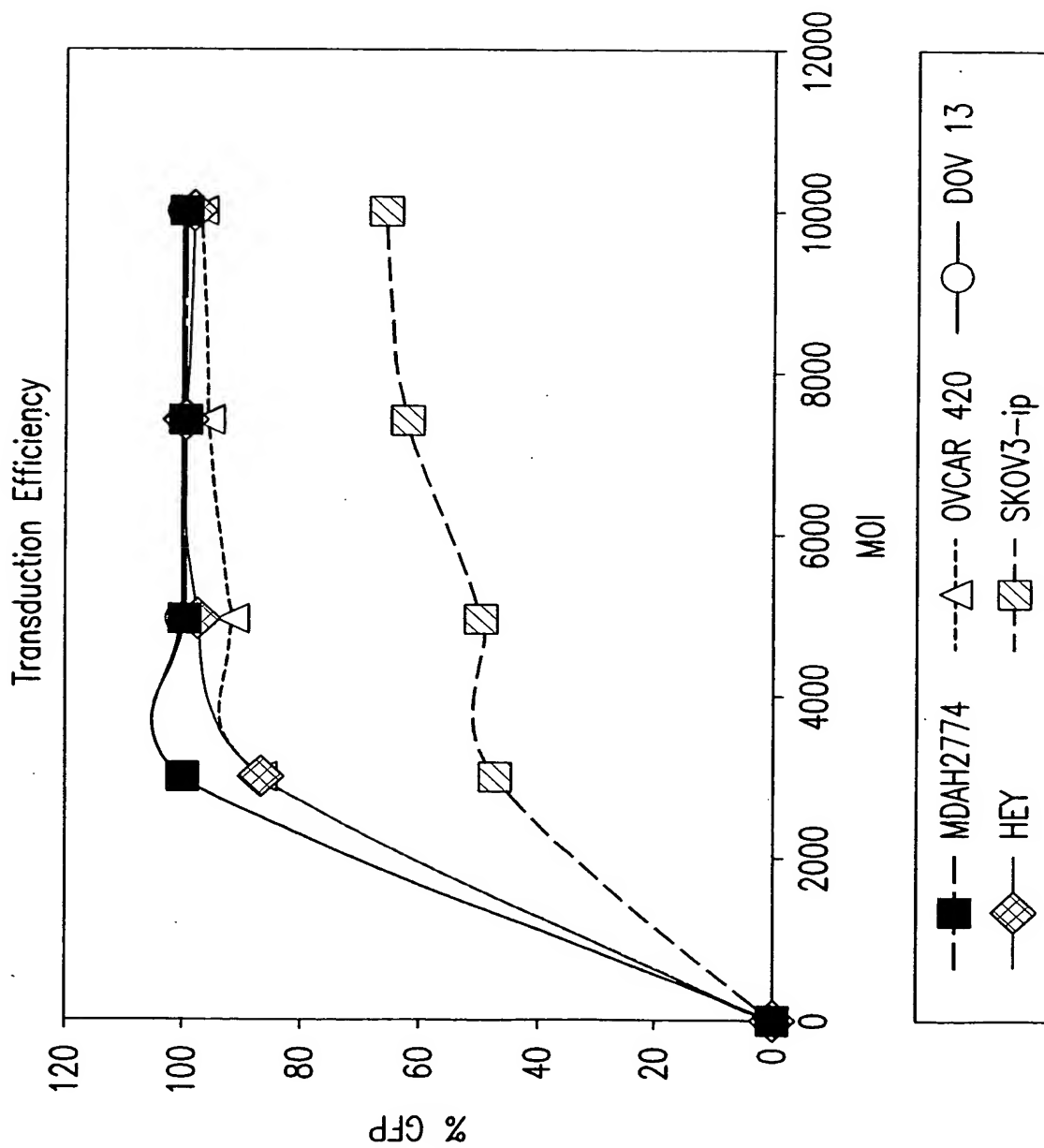


FIG.50

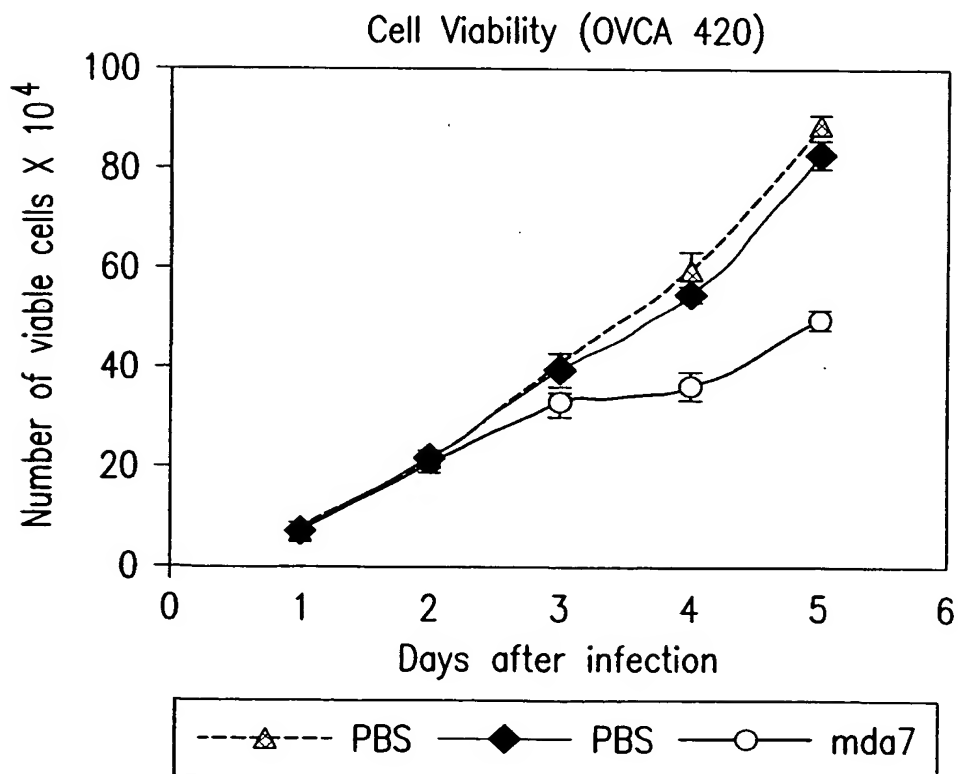
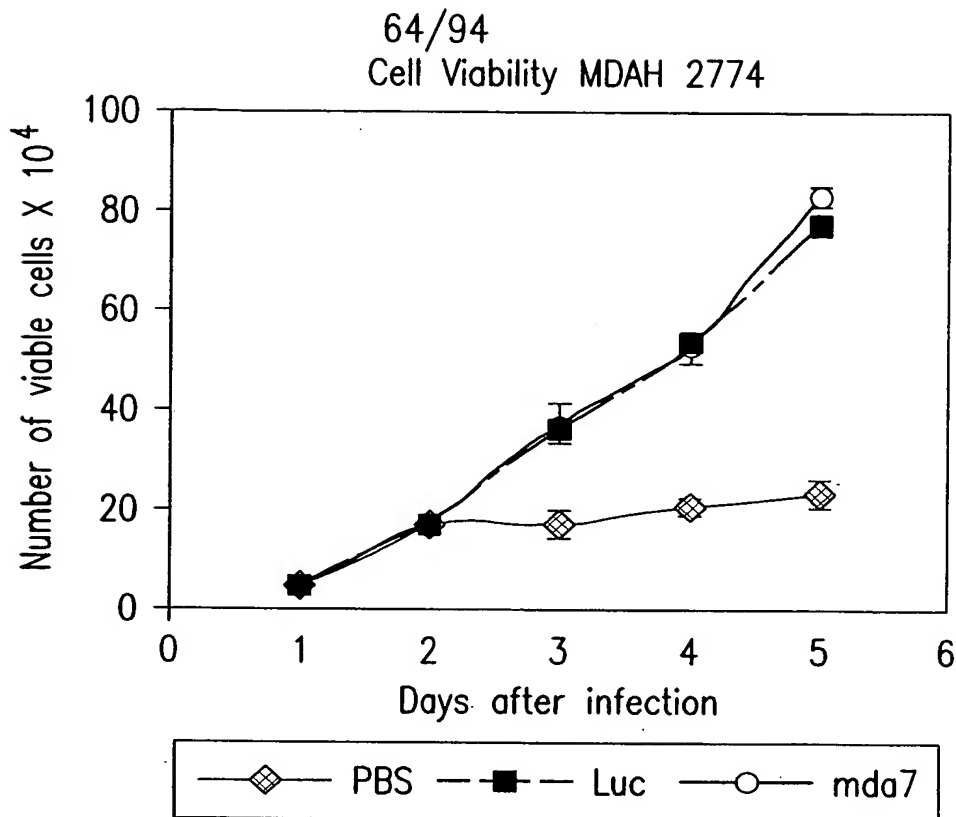


FIG.51



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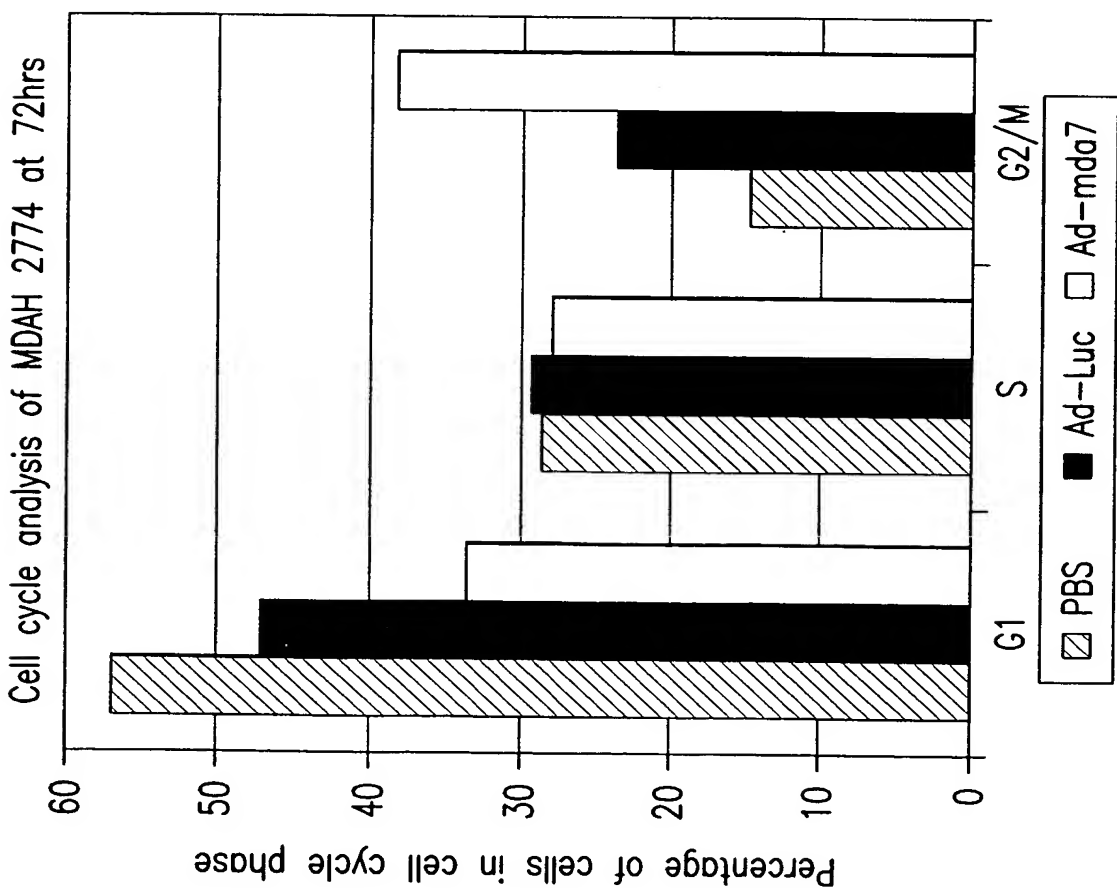
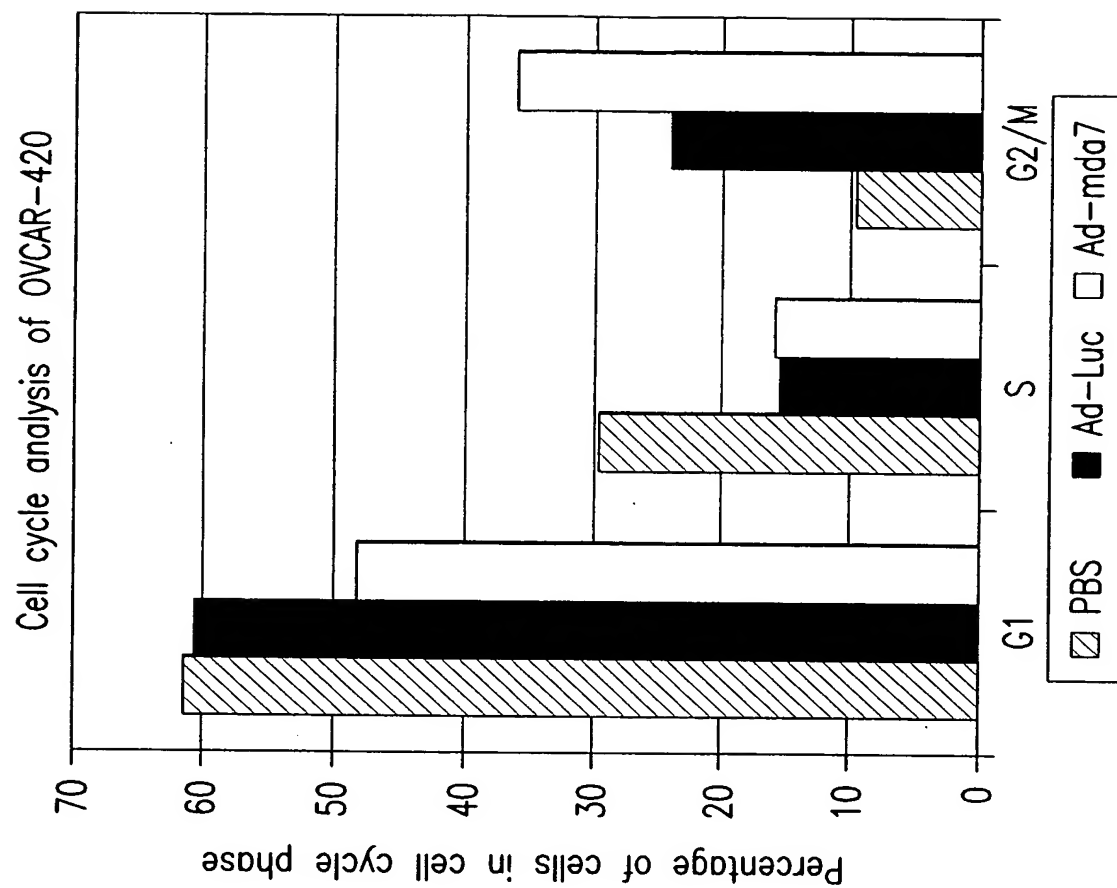


FIG.52

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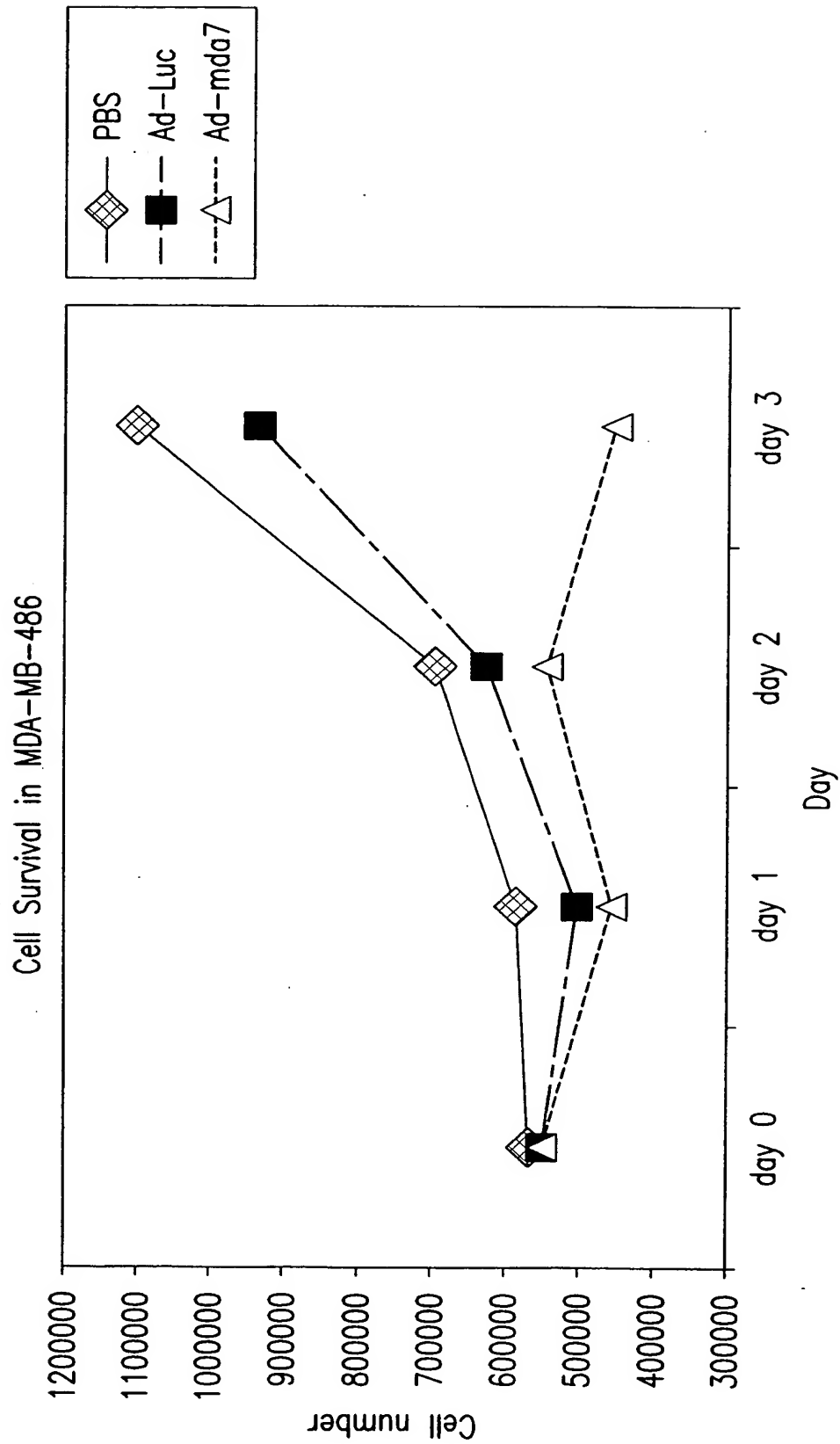


FIG.53

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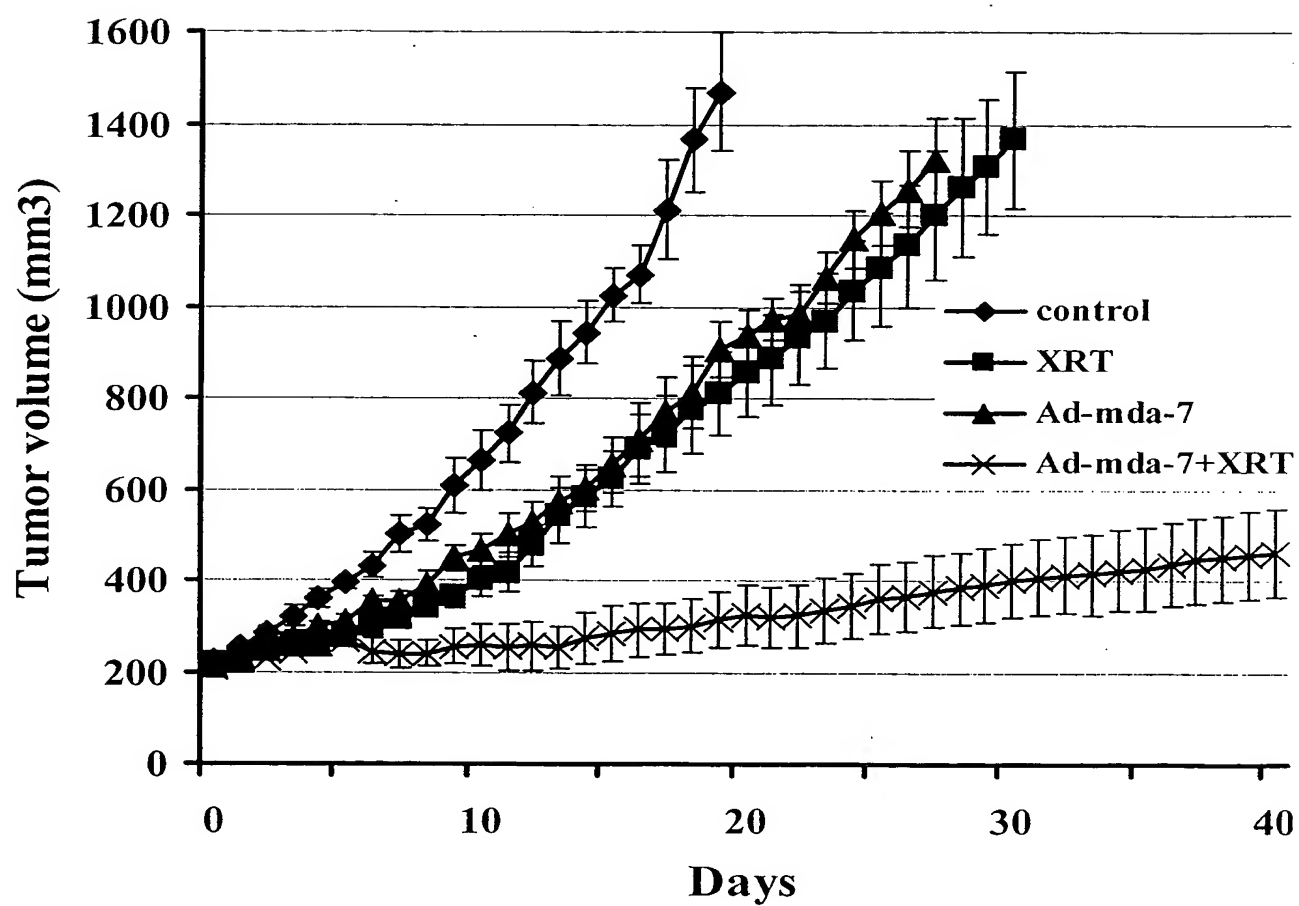


FIG.54A

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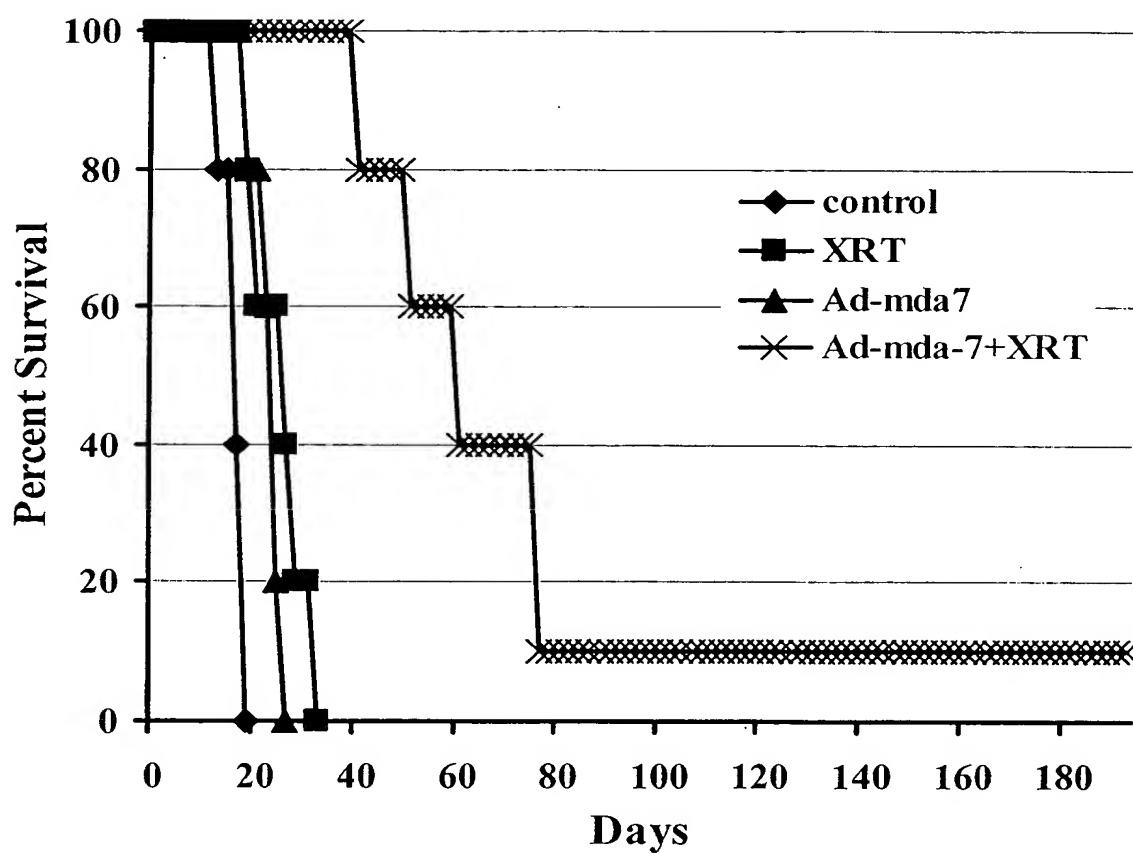


FIG.54B

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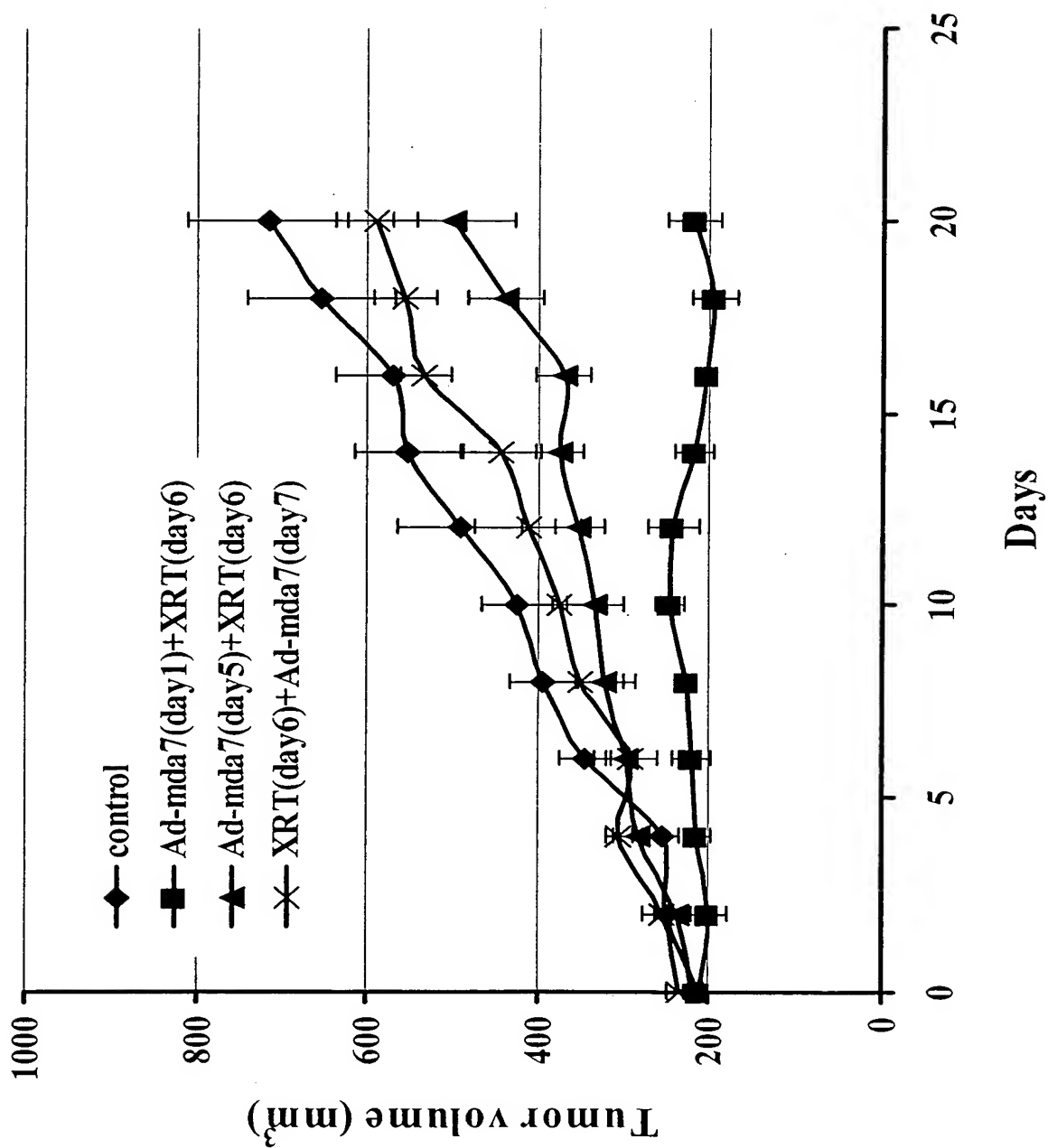


FIG.55

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# Apoptotic Index

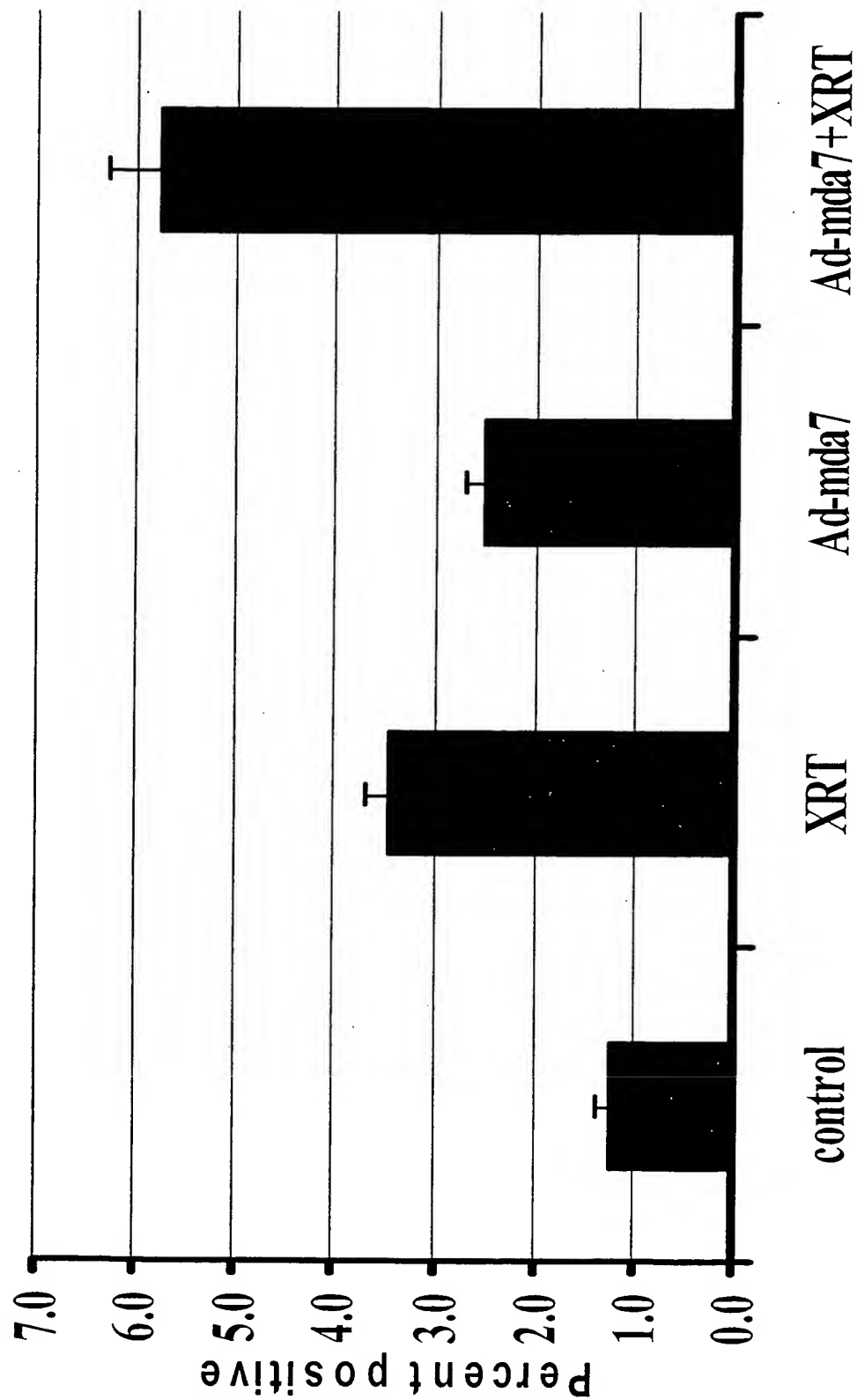


FIG. 56

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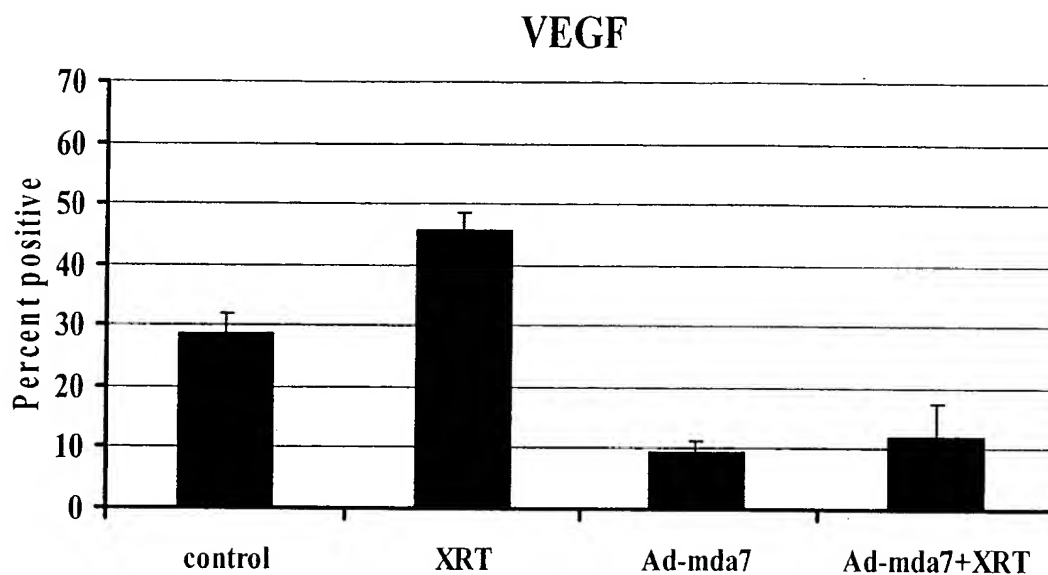


FIG.57A

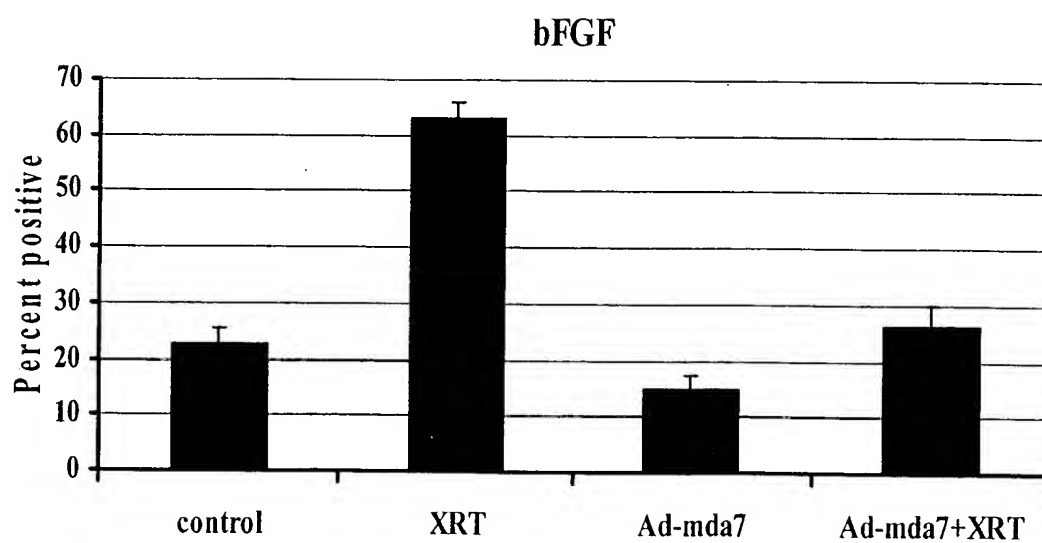


FIG.57B

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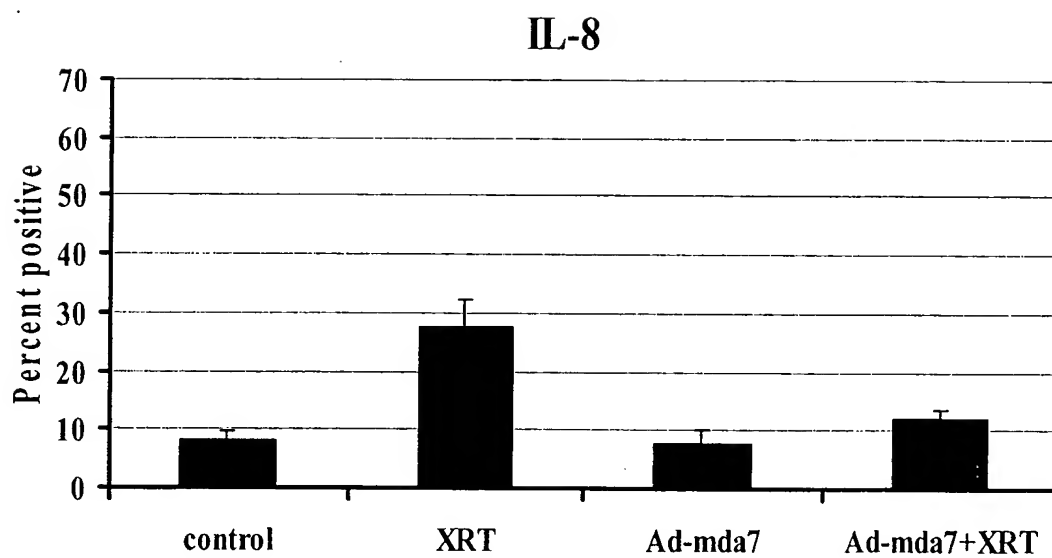


FIG.57C



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# Micro Vessel Density

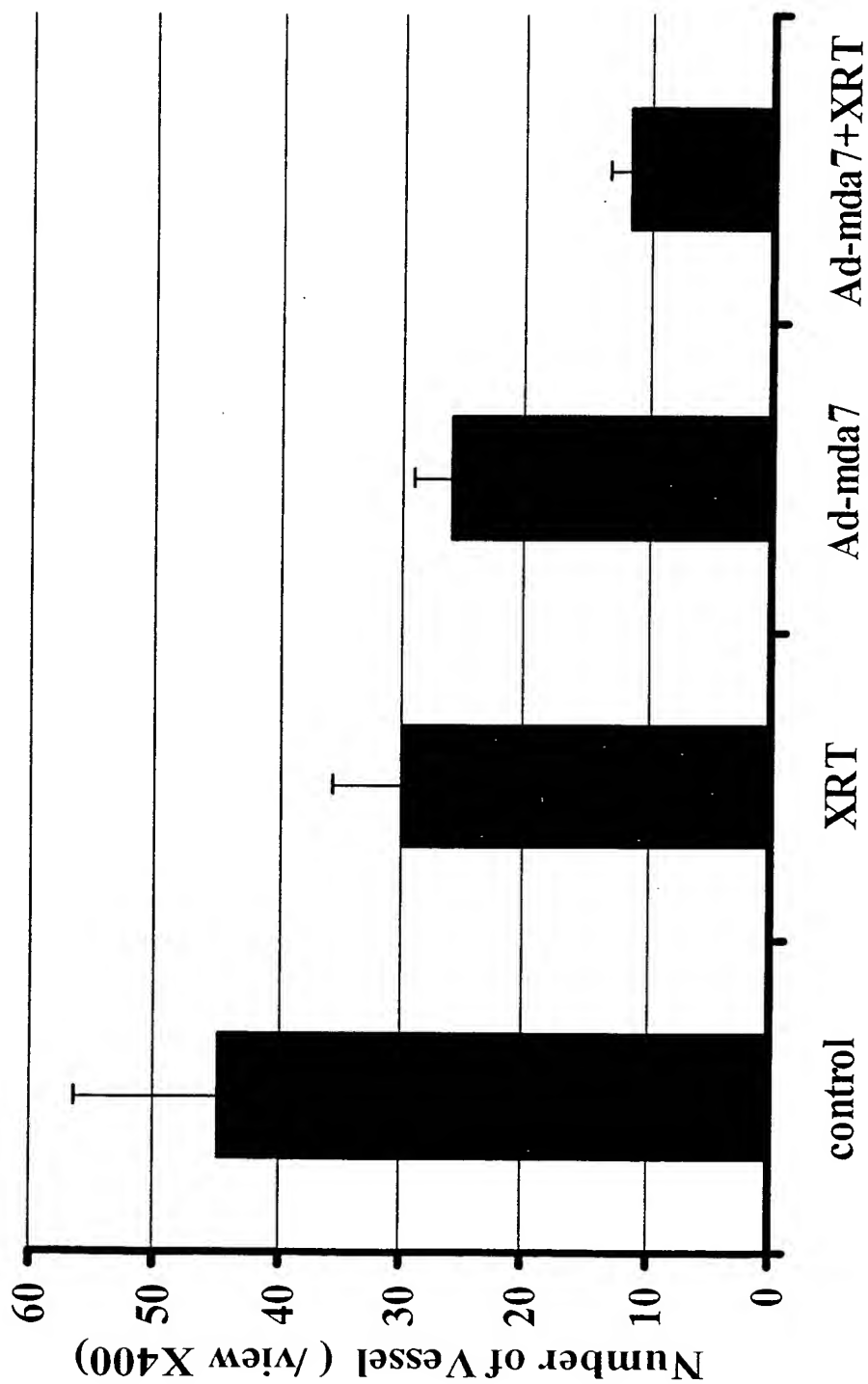


FIG.58

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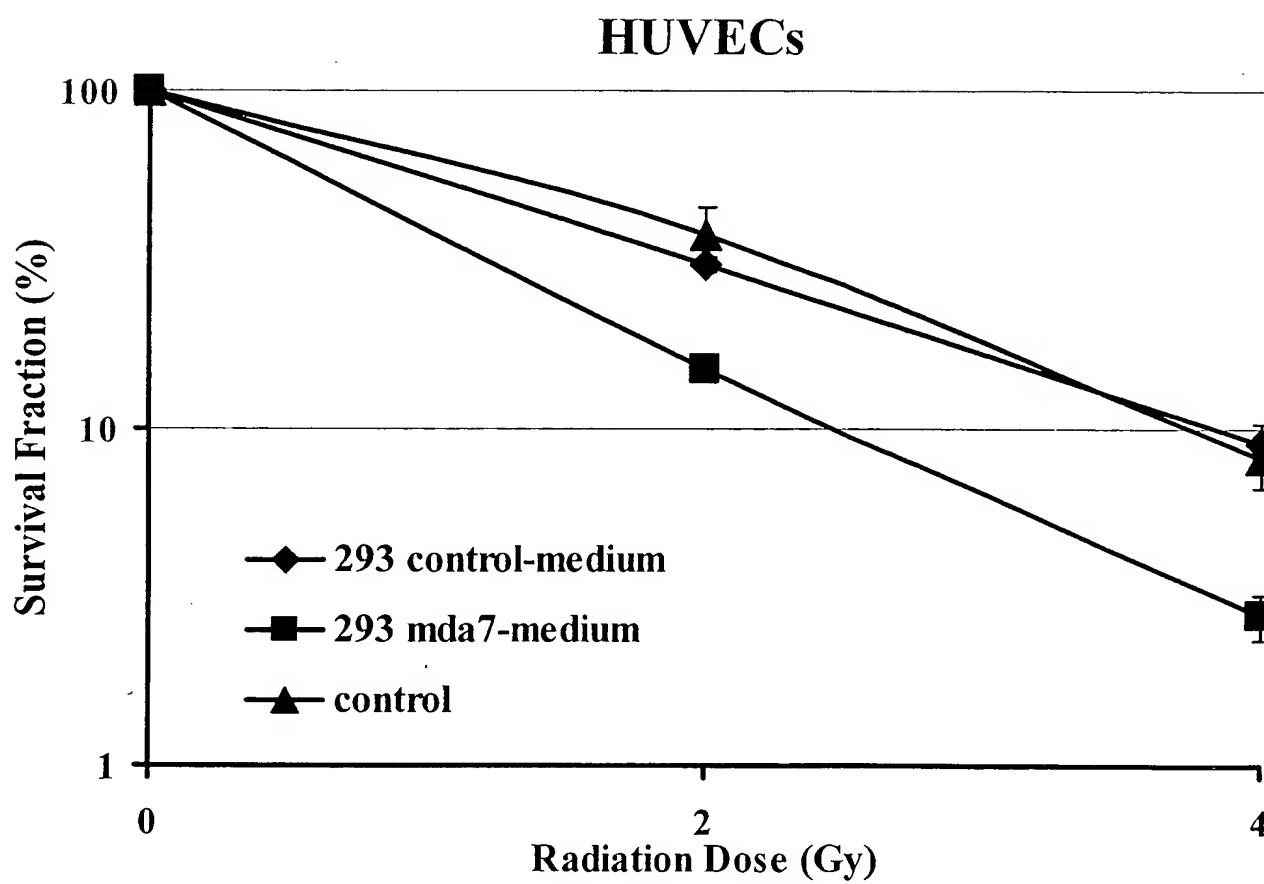


FIG.59A

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# HUVECs

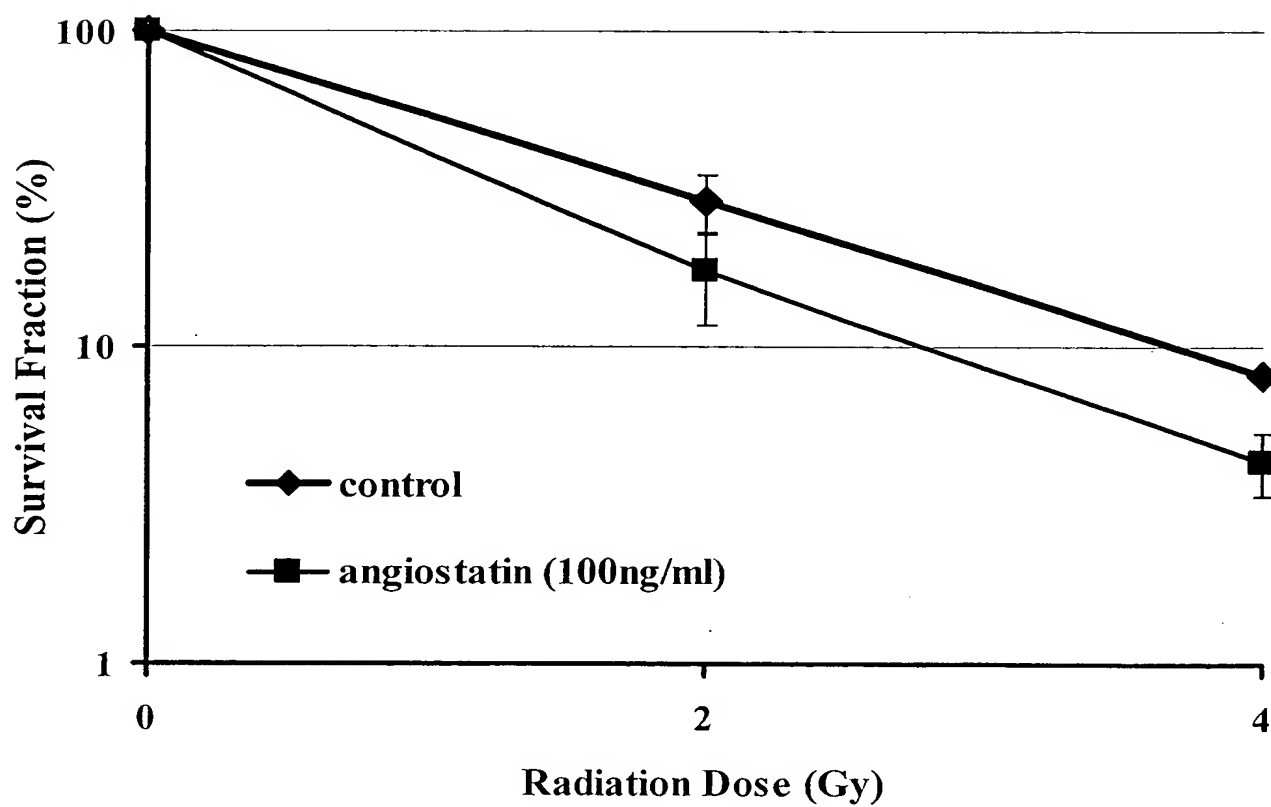


FIG.59B

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# HUVECs

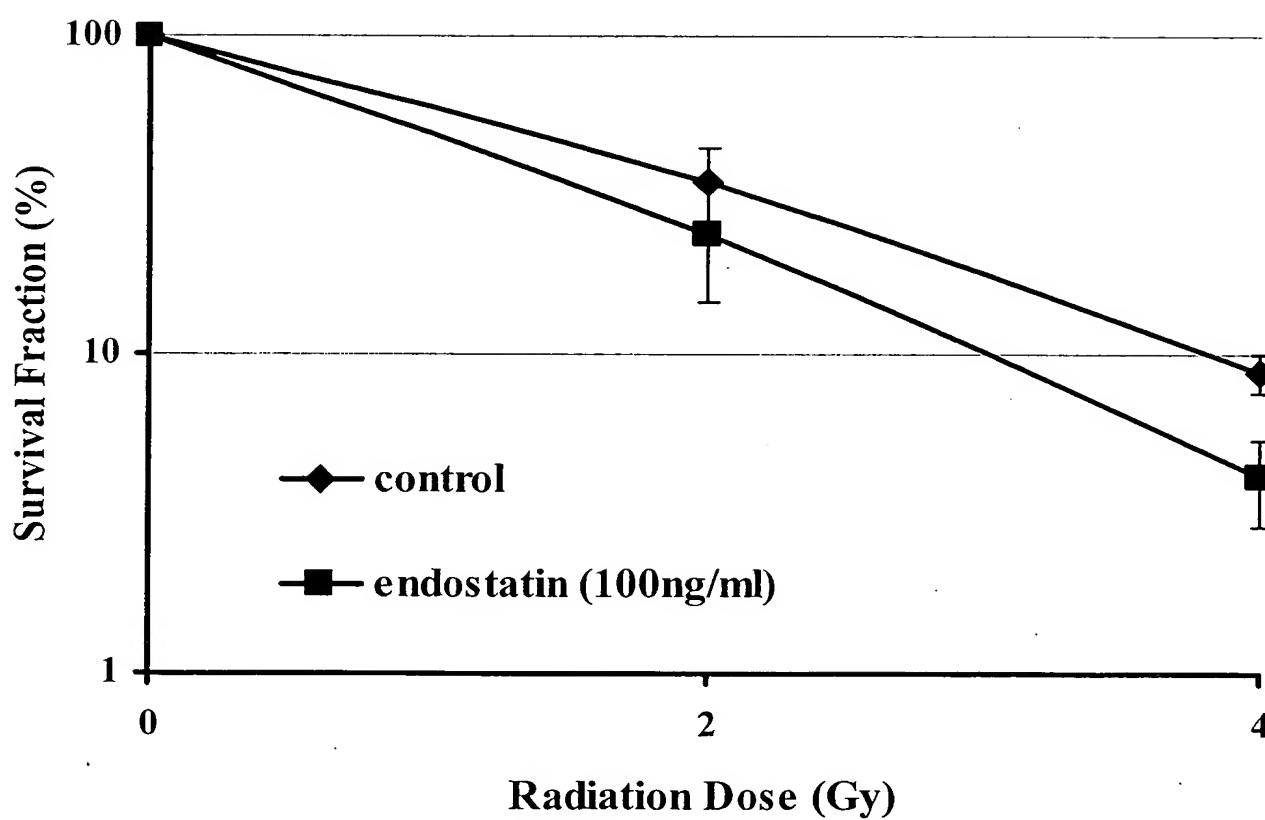


FIG.59C

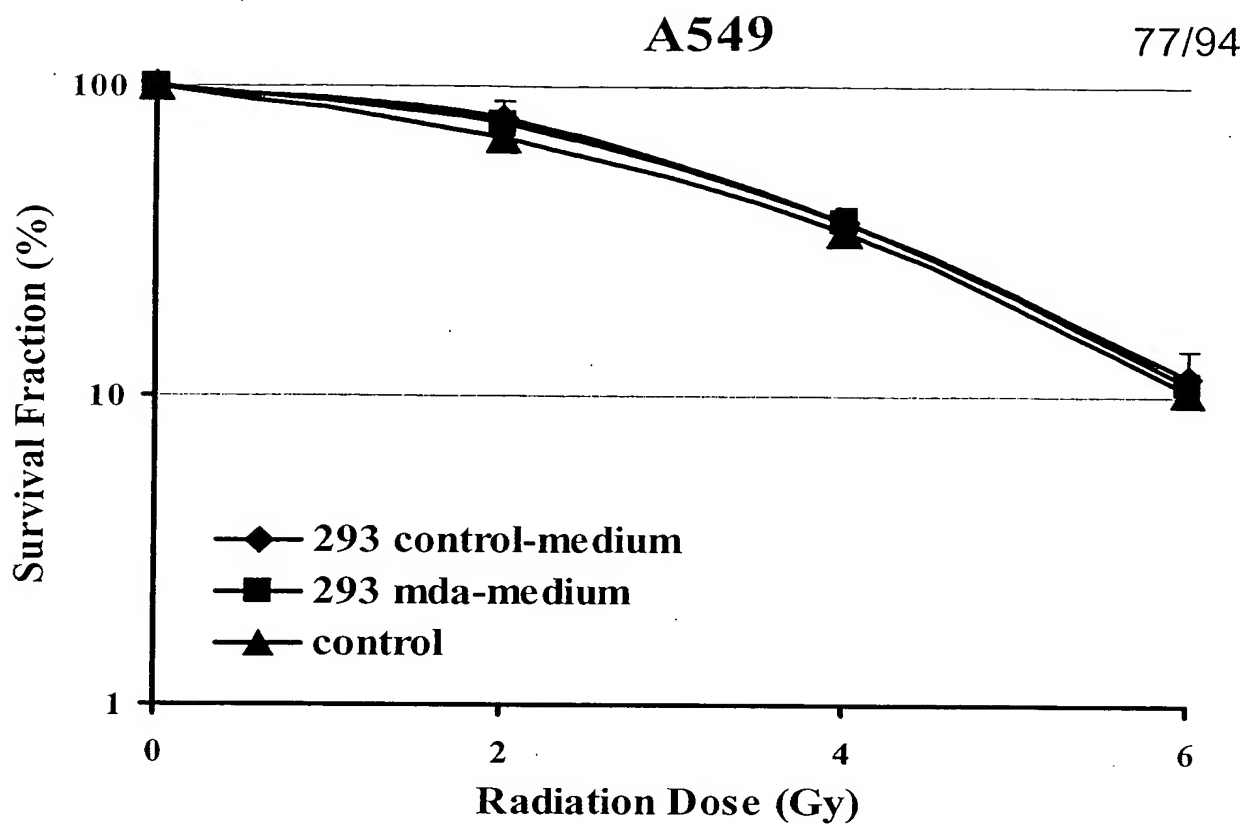


FIG. 60A

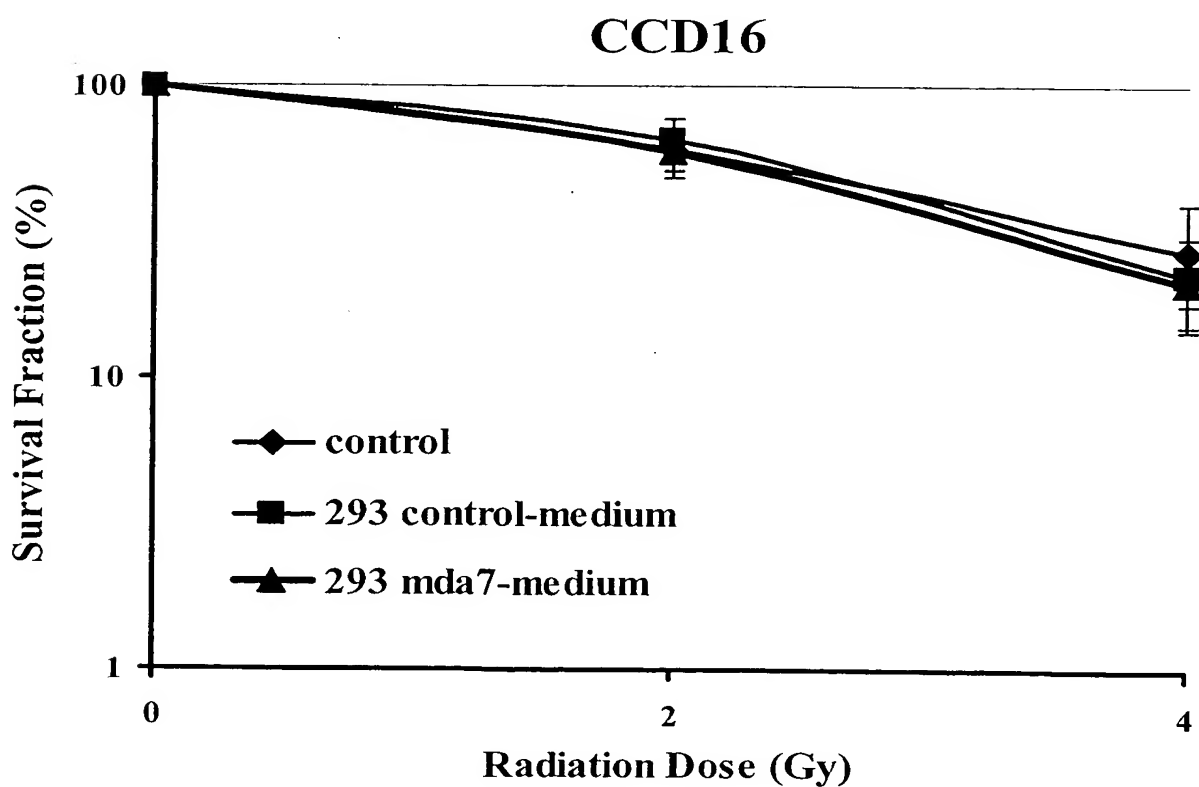


FIG. 60B

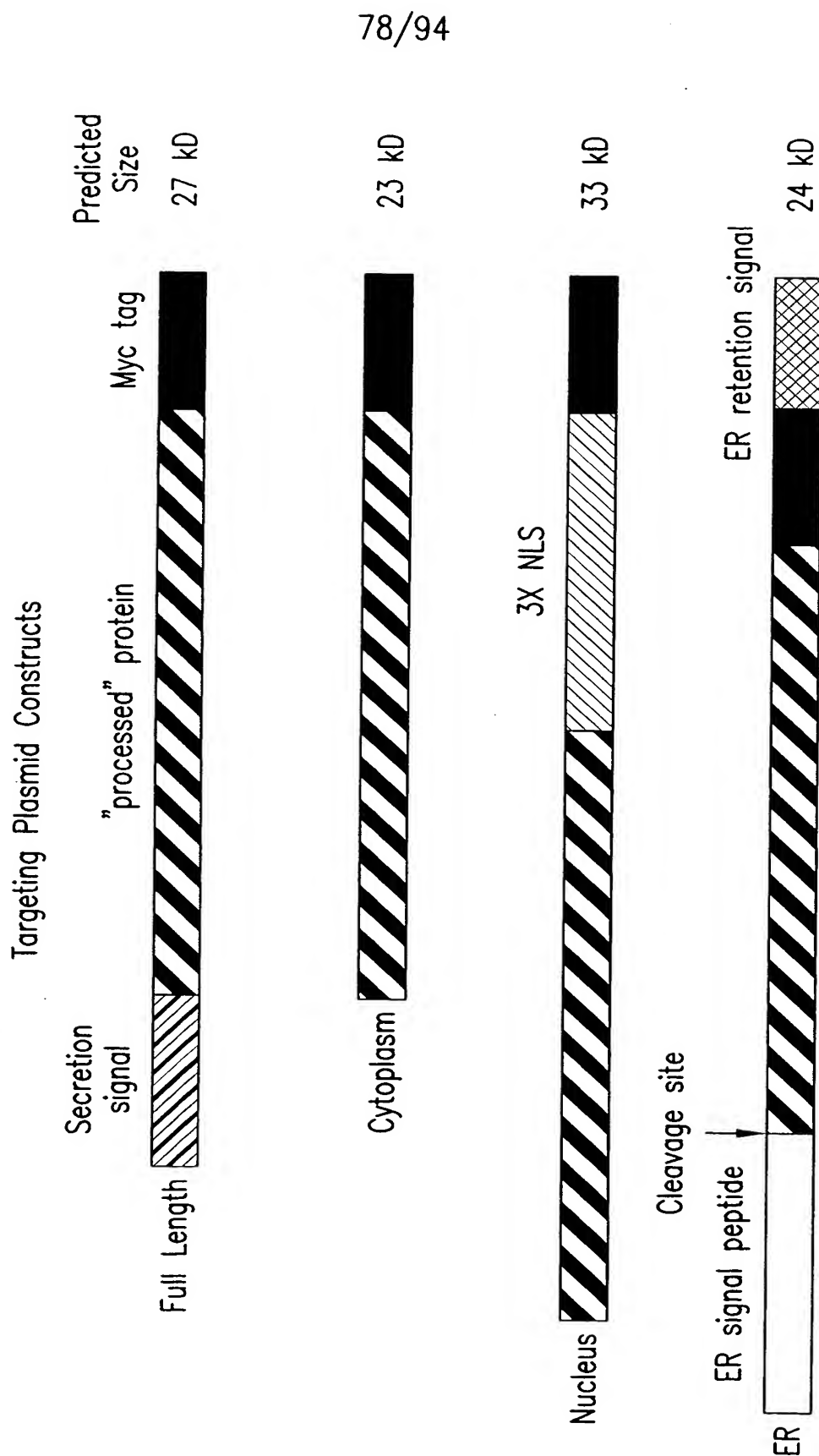


FIG. 61

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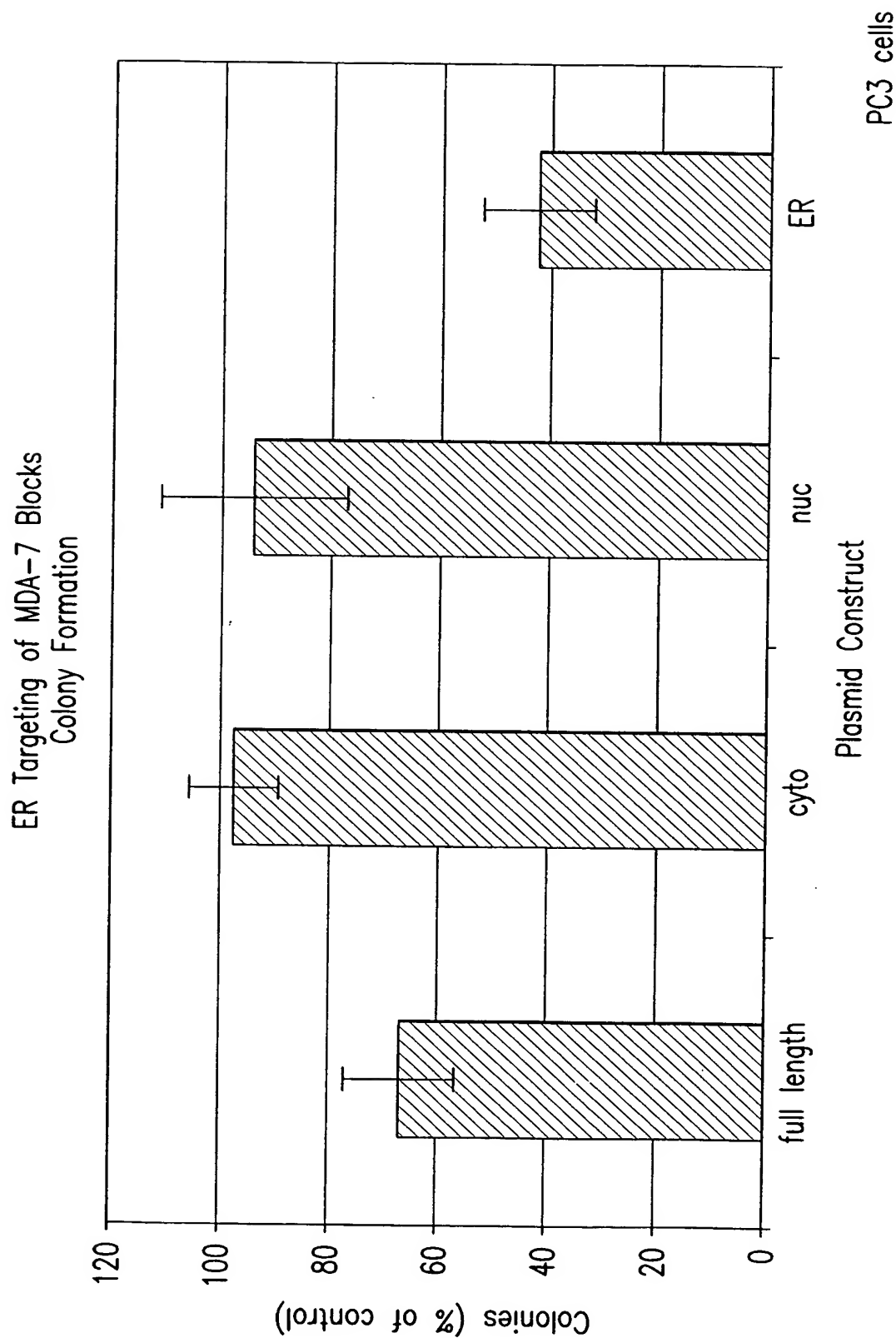


FIG.62

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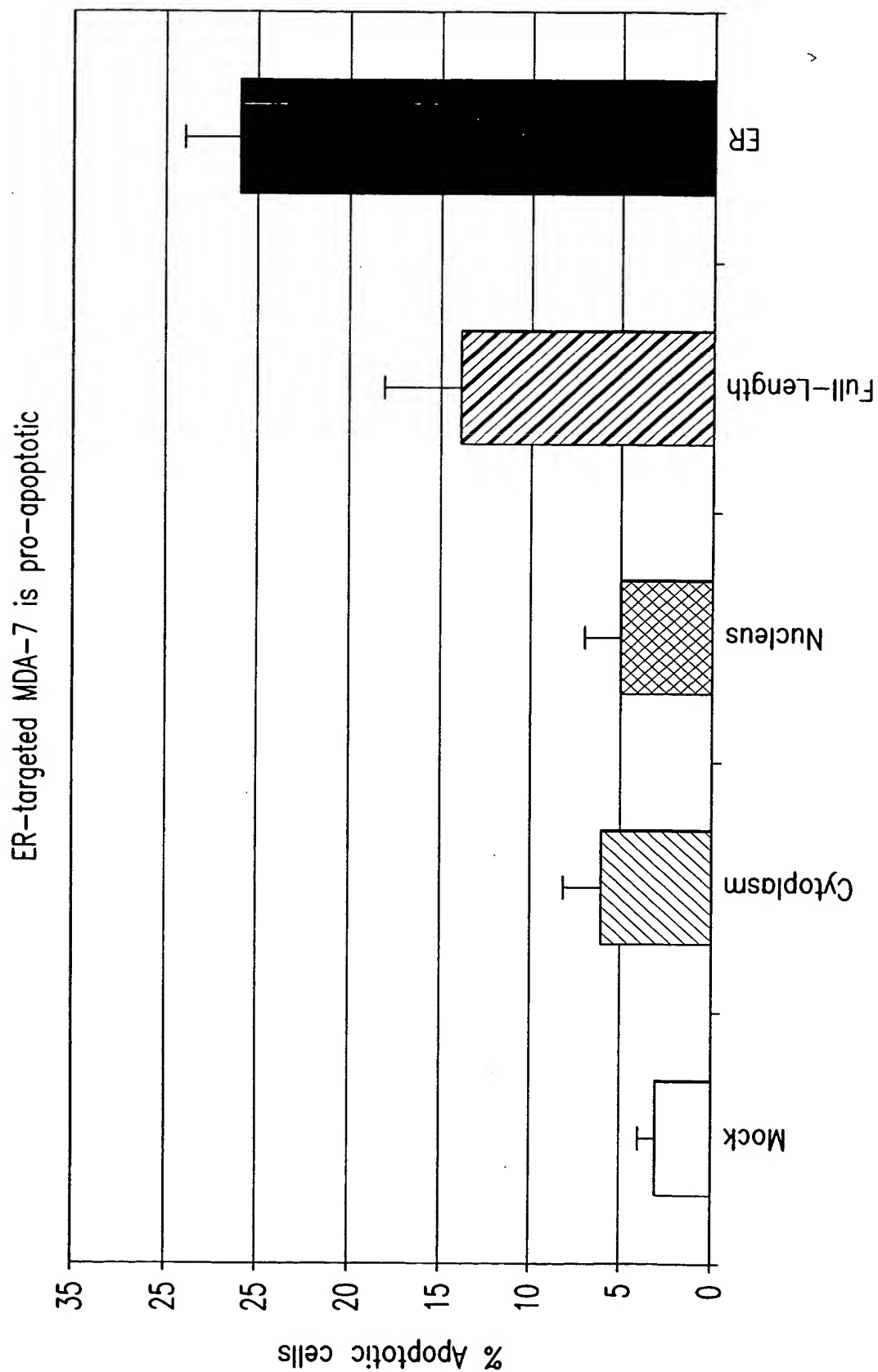


FIG. 63



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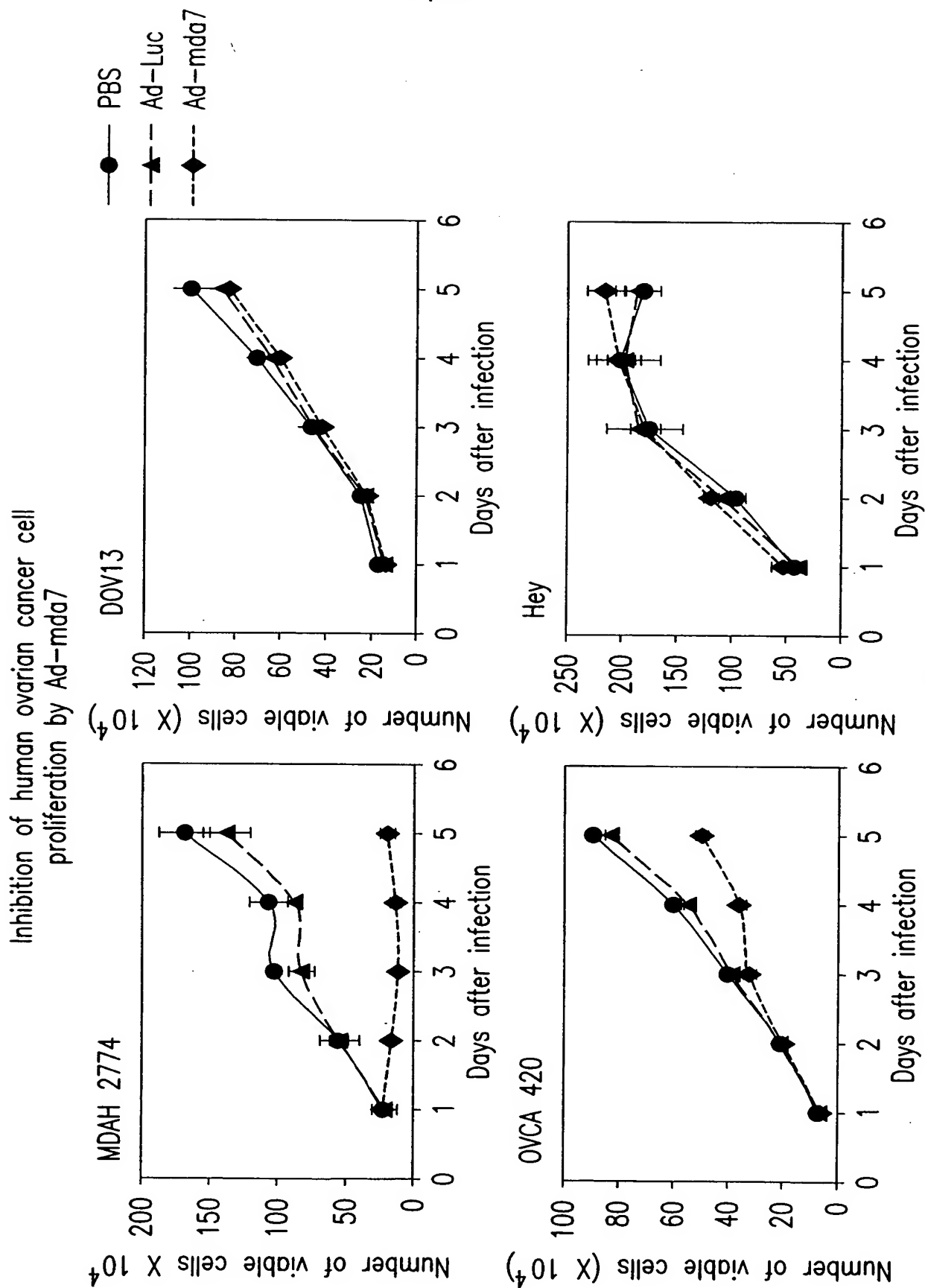


FIG.64

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Induction of cell cycle arrest by MDA-7  
(MDAH 2774)

|               | G1    | S     | G2M   | Apoptosis<br>Sub G0 |
|---------------|-------|-------|-------|---------------------|
| 24 hrs<br>PBS | 30.55 | 34.55 | 34.9  | 2.05                |
| Ad.Luc        | 32.95 | 36.6  | 30.45 | 1.41                |
| Ad.mda-7      | 27.45 | 29.75 | 42.85 | 4                   |
| 48 hrs<br>PBS | 40.8  | 32.6  | 26.6  | 0.955               |
| Ad.Luc        | 28.75 | 32.45 | 38.8  | 0.84                |
| Ad.mda-7      | 18.3  | 78    | 54.5  | 10.36               |

FIG.65A

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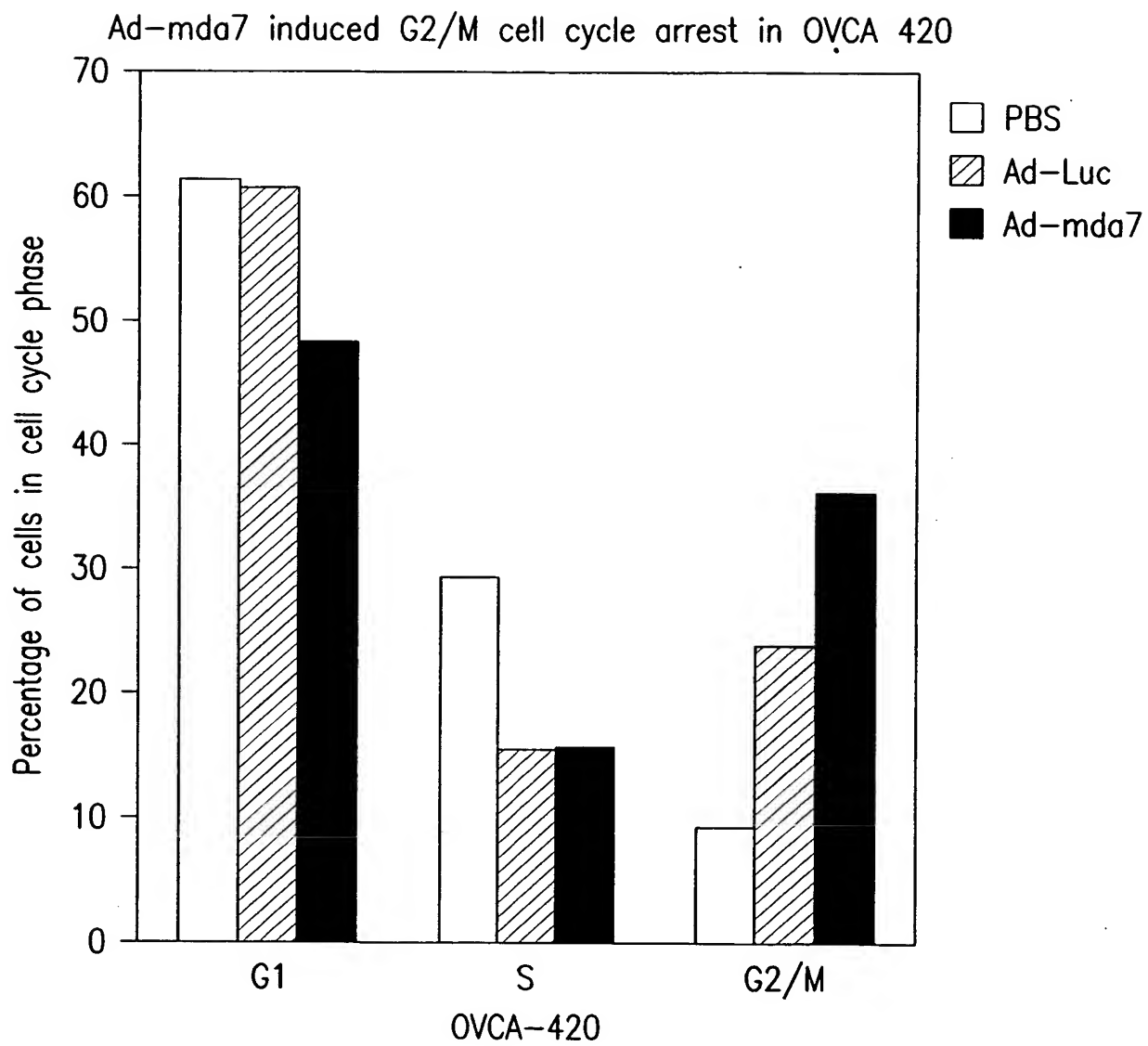


FIG.65B

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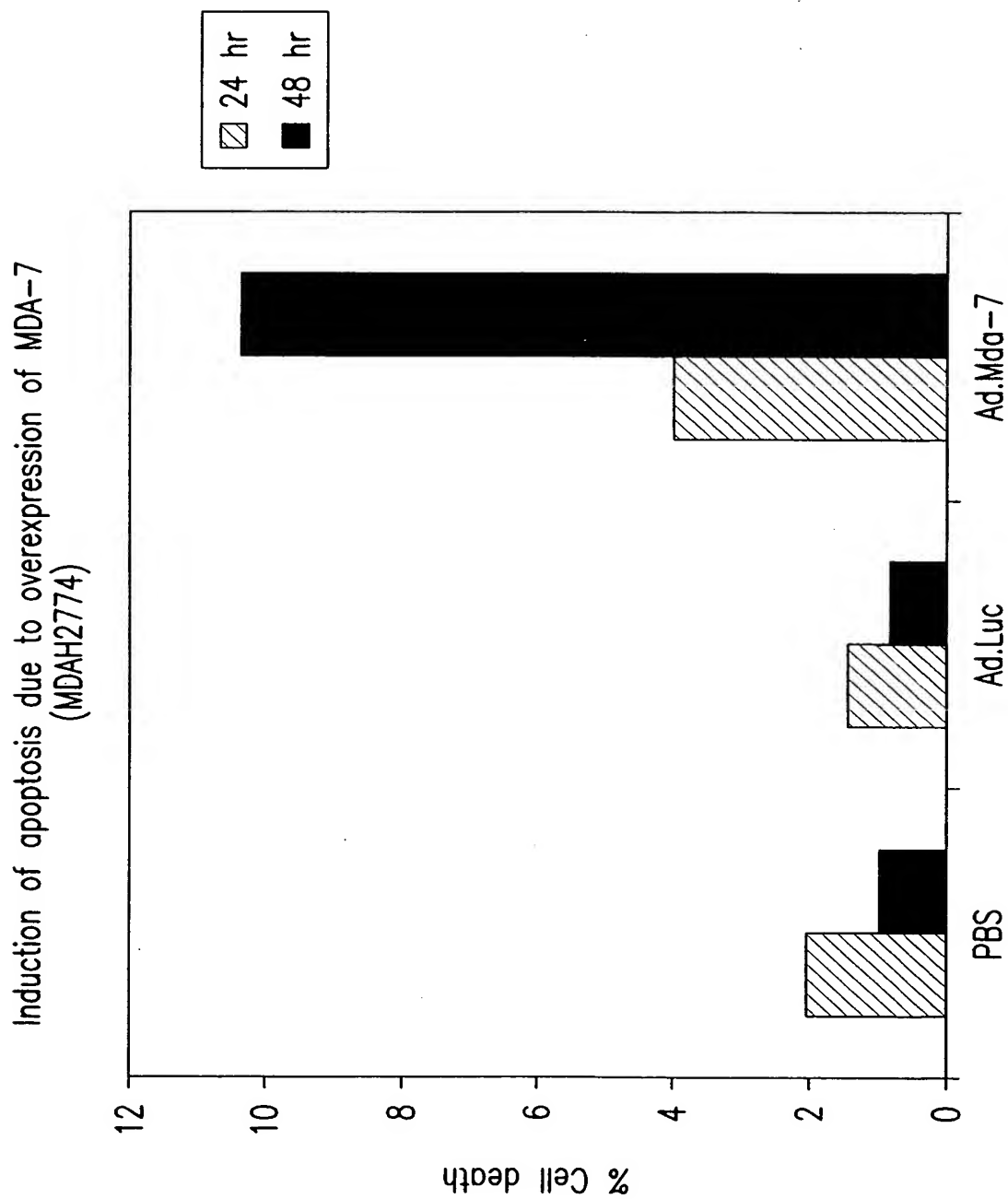


FIG. 66

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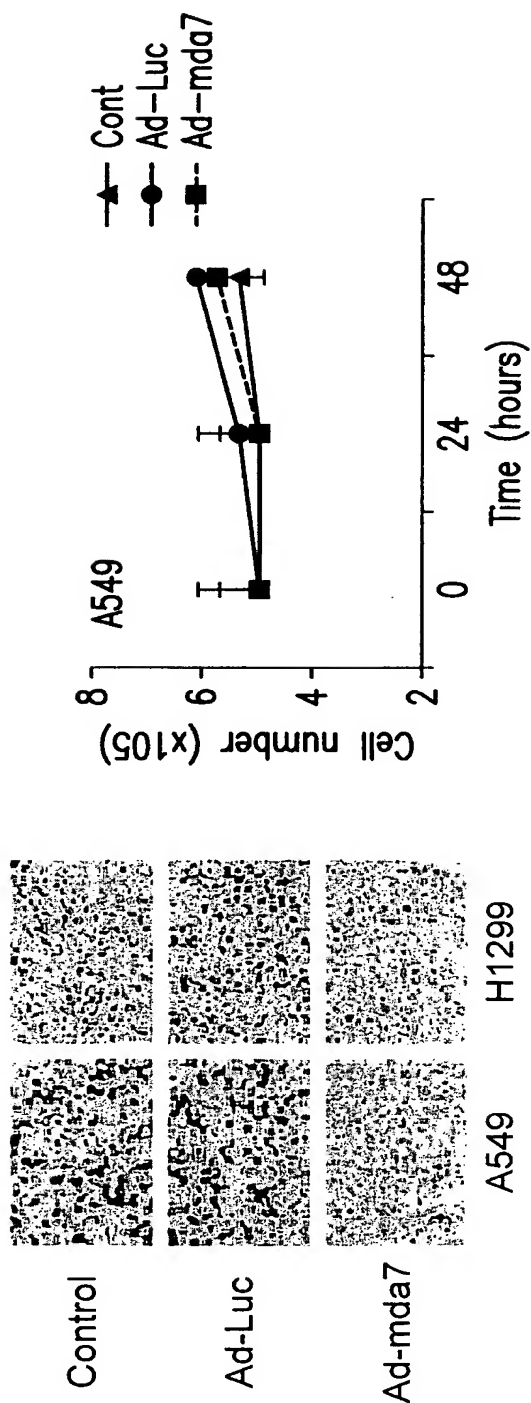


FIG.67A

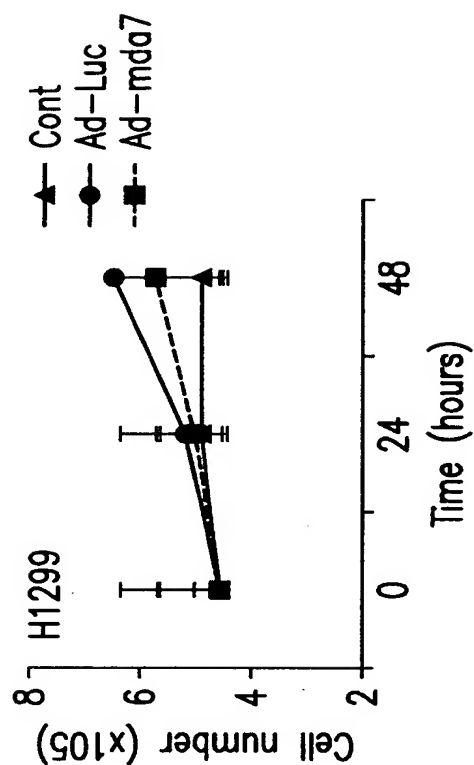


FIG.67B

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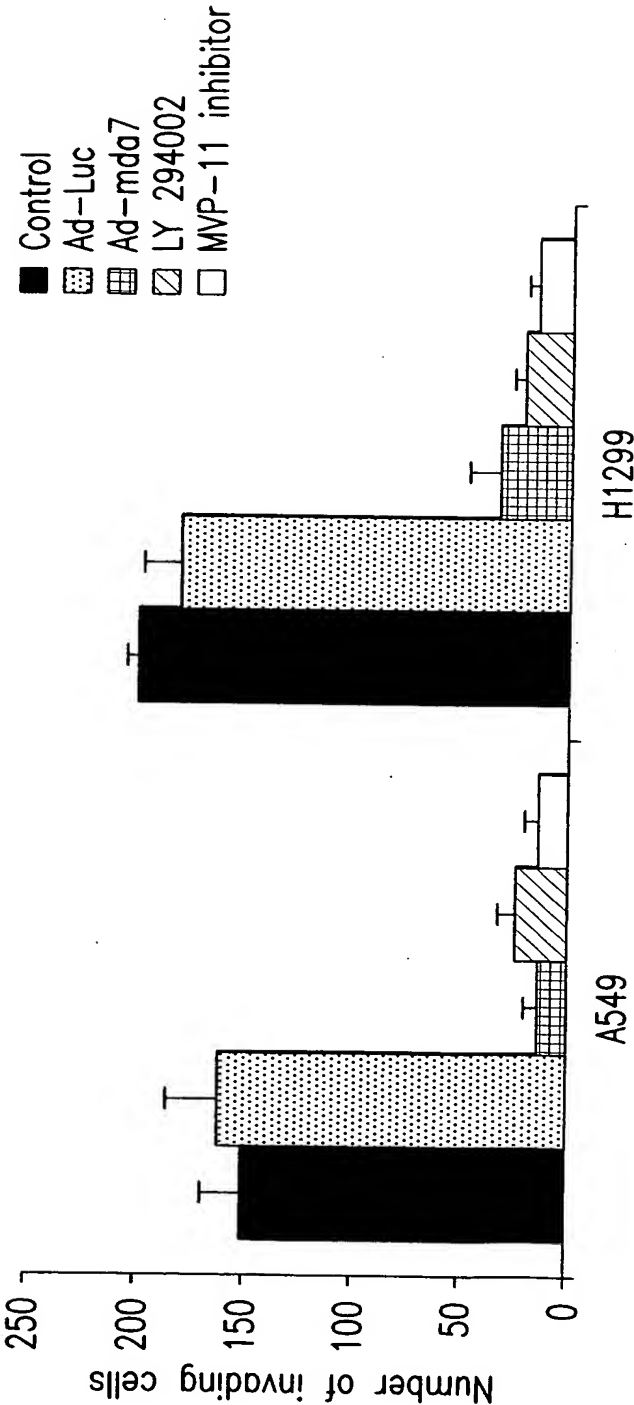


FIG.68

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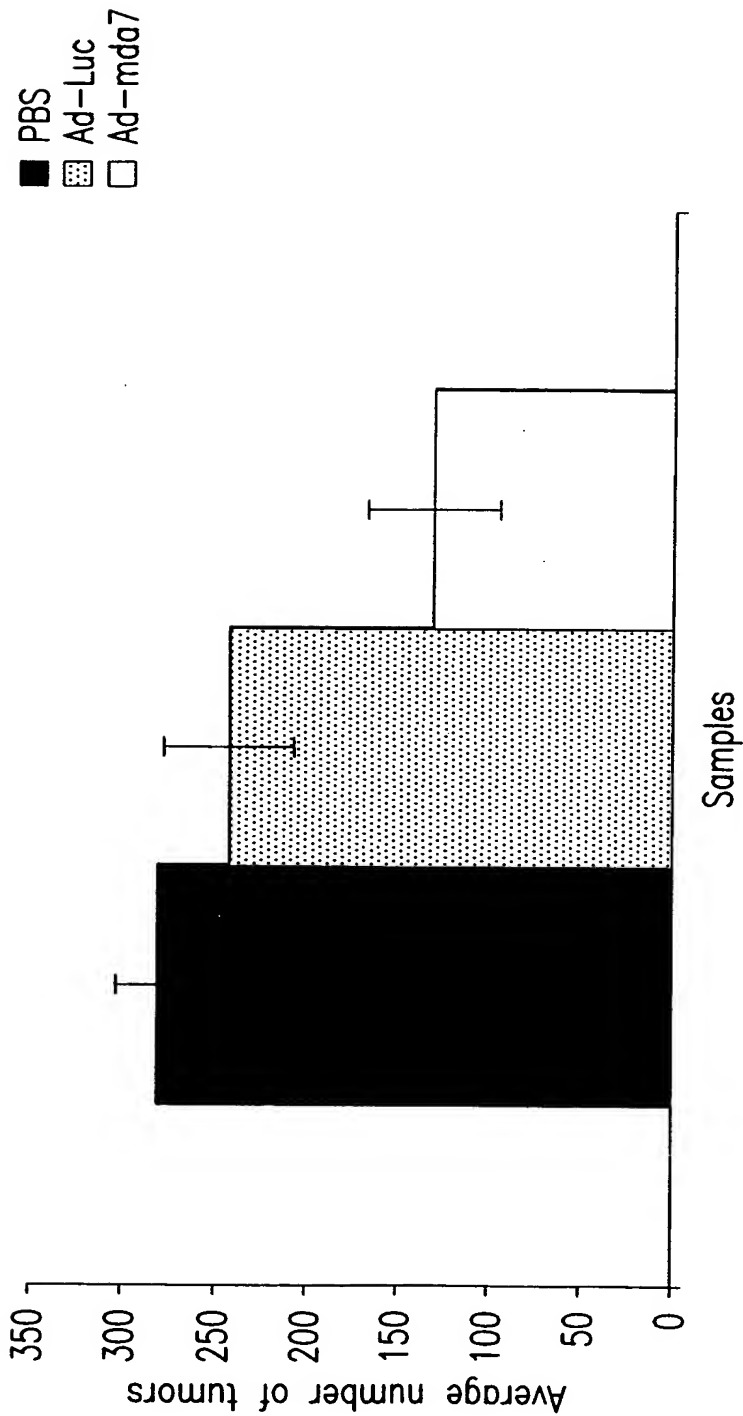


FIG.69

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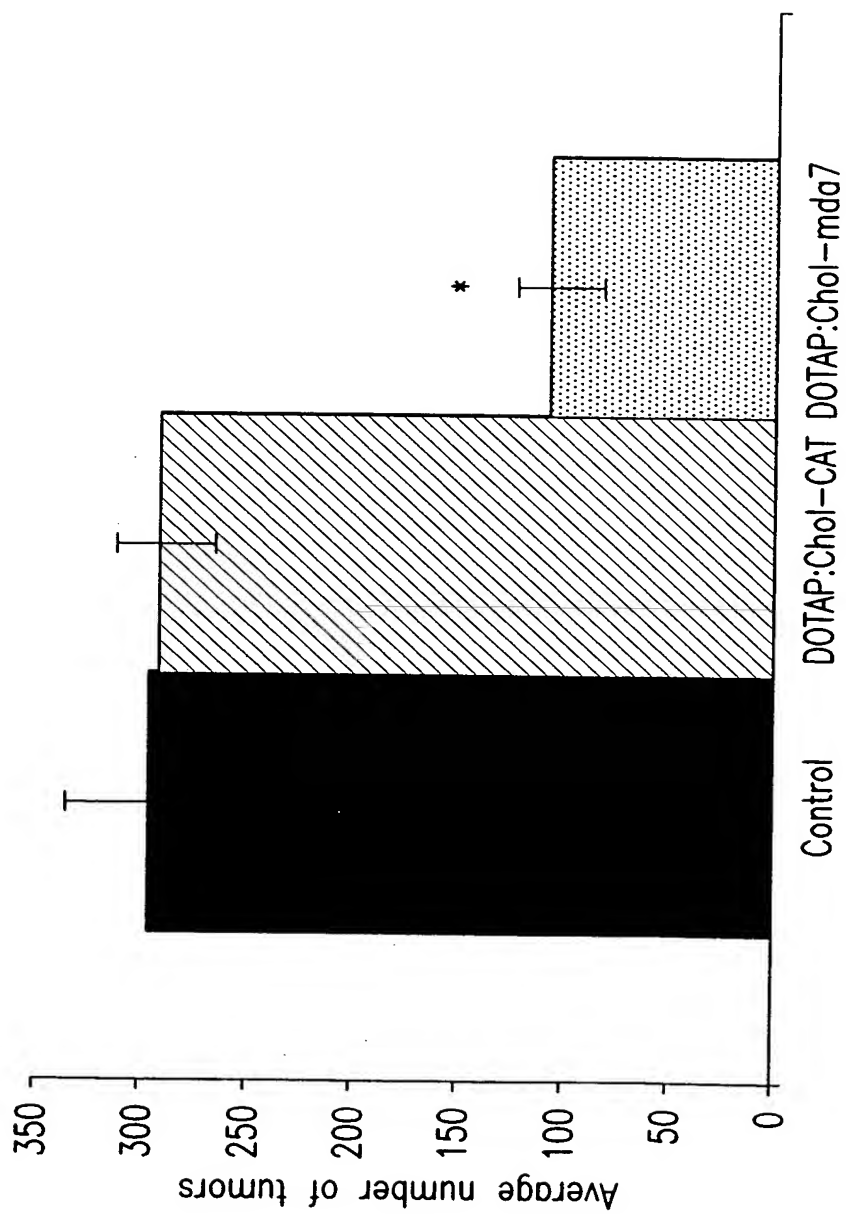


FIG. 70



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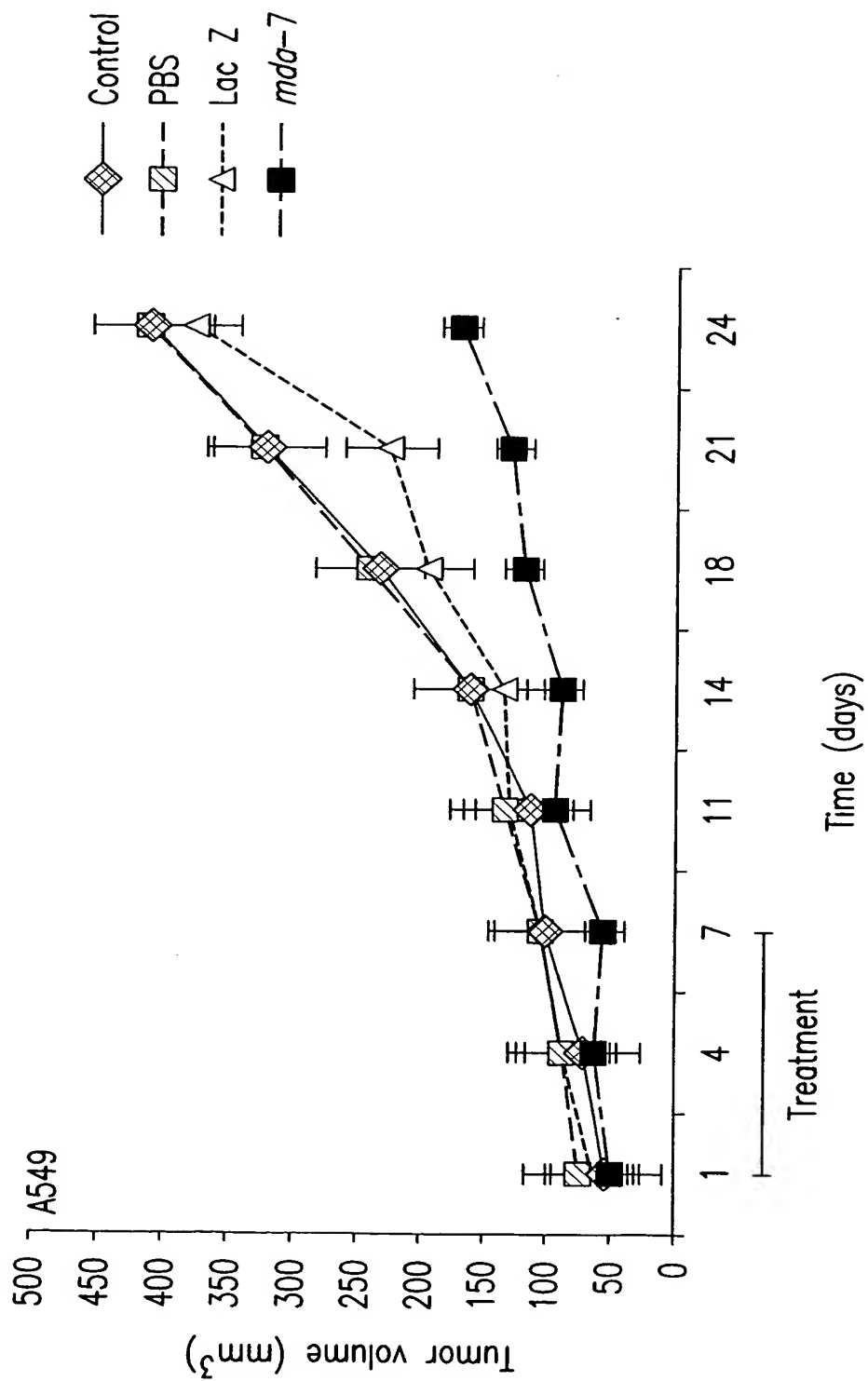


FIG.71A

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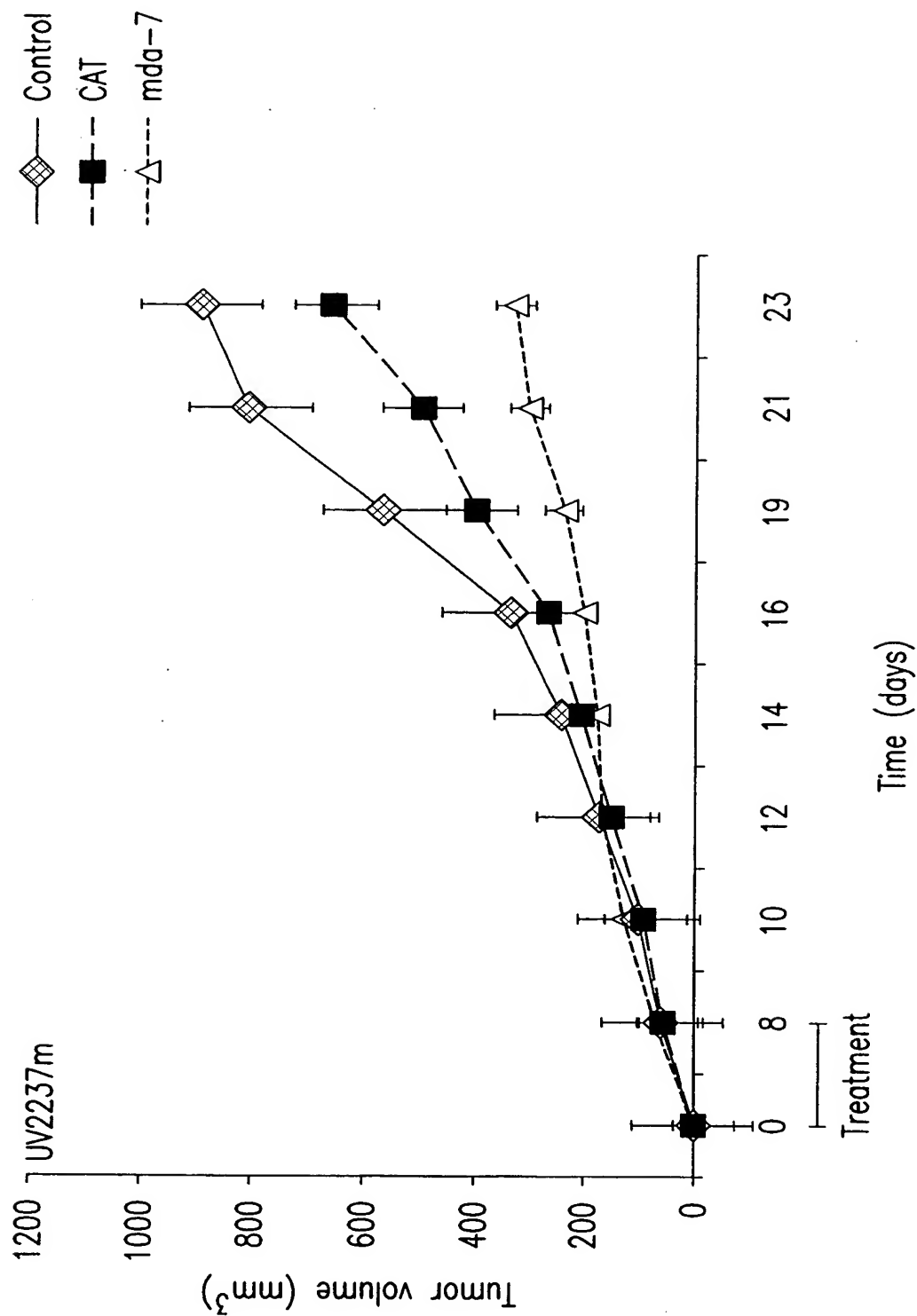


FIG.71B

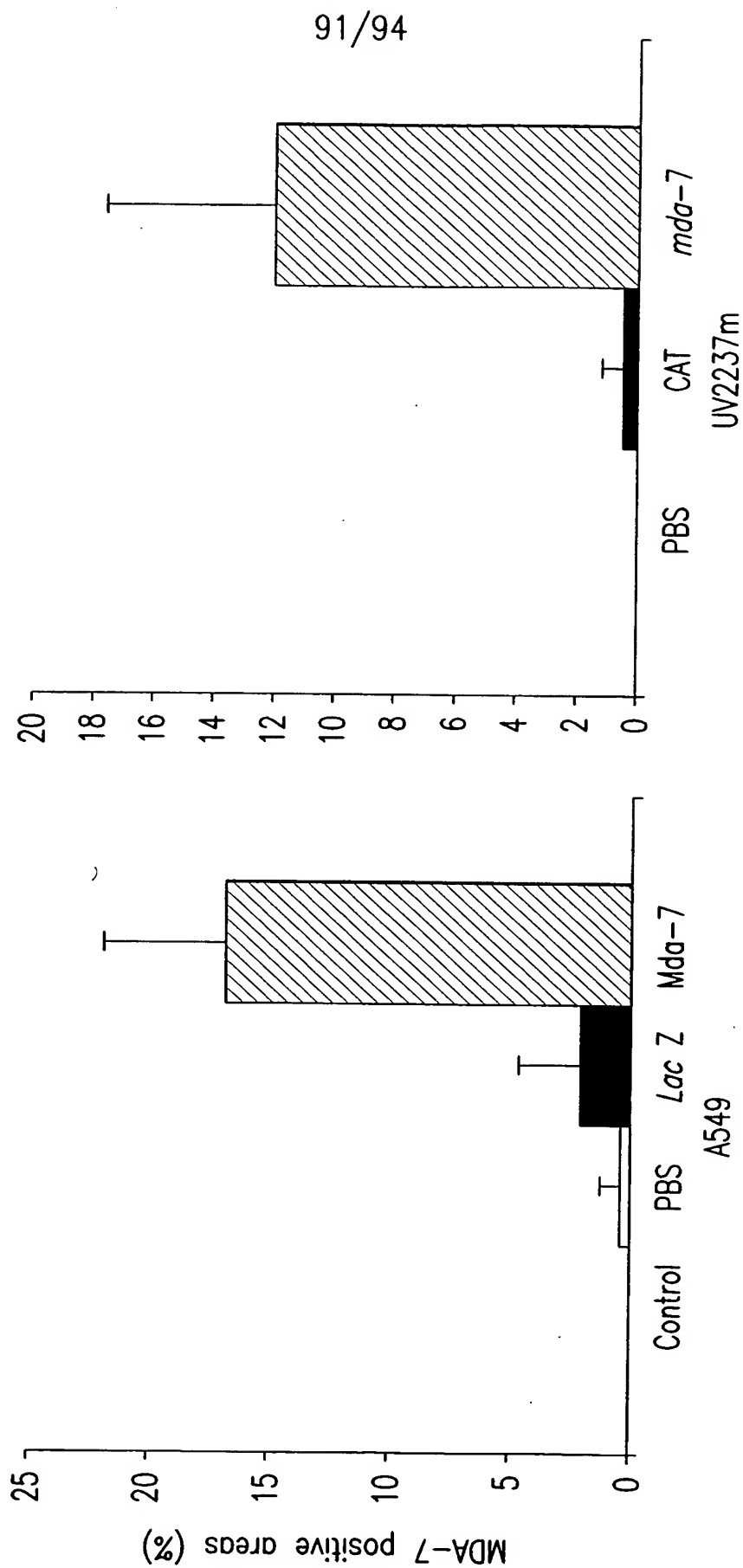


FIG.71C

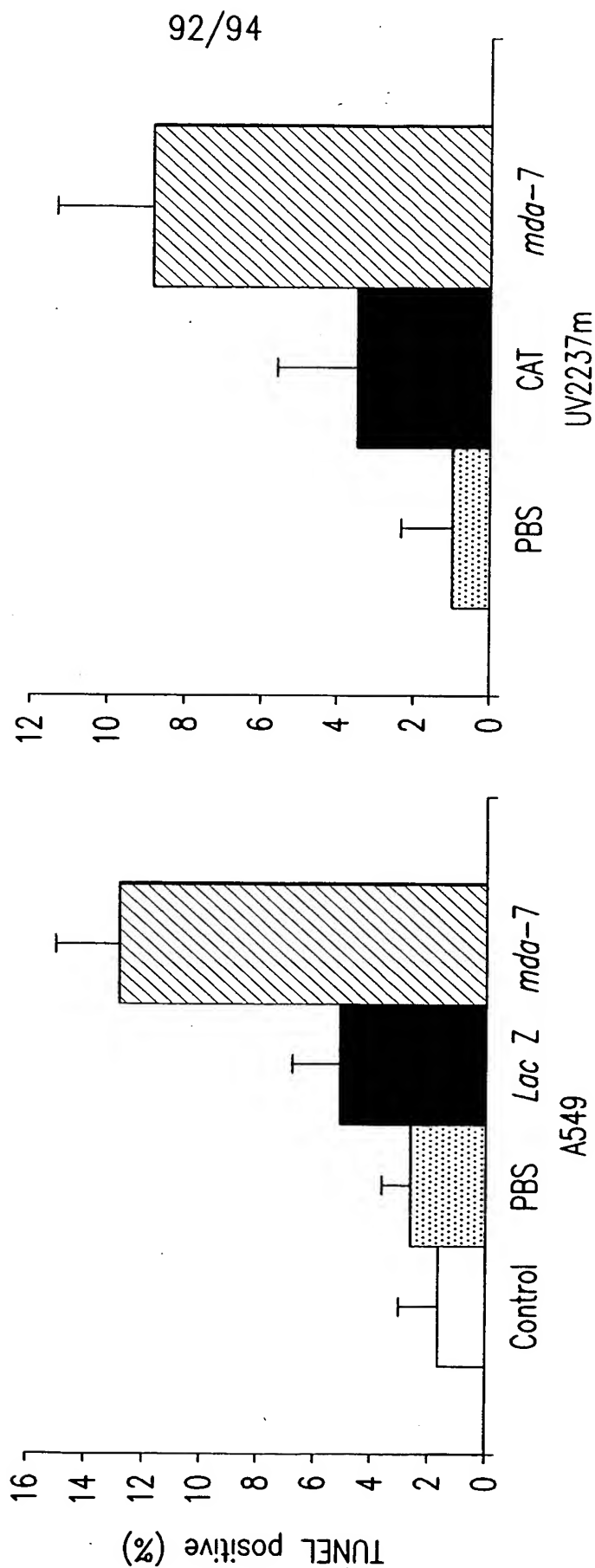


FIG.72

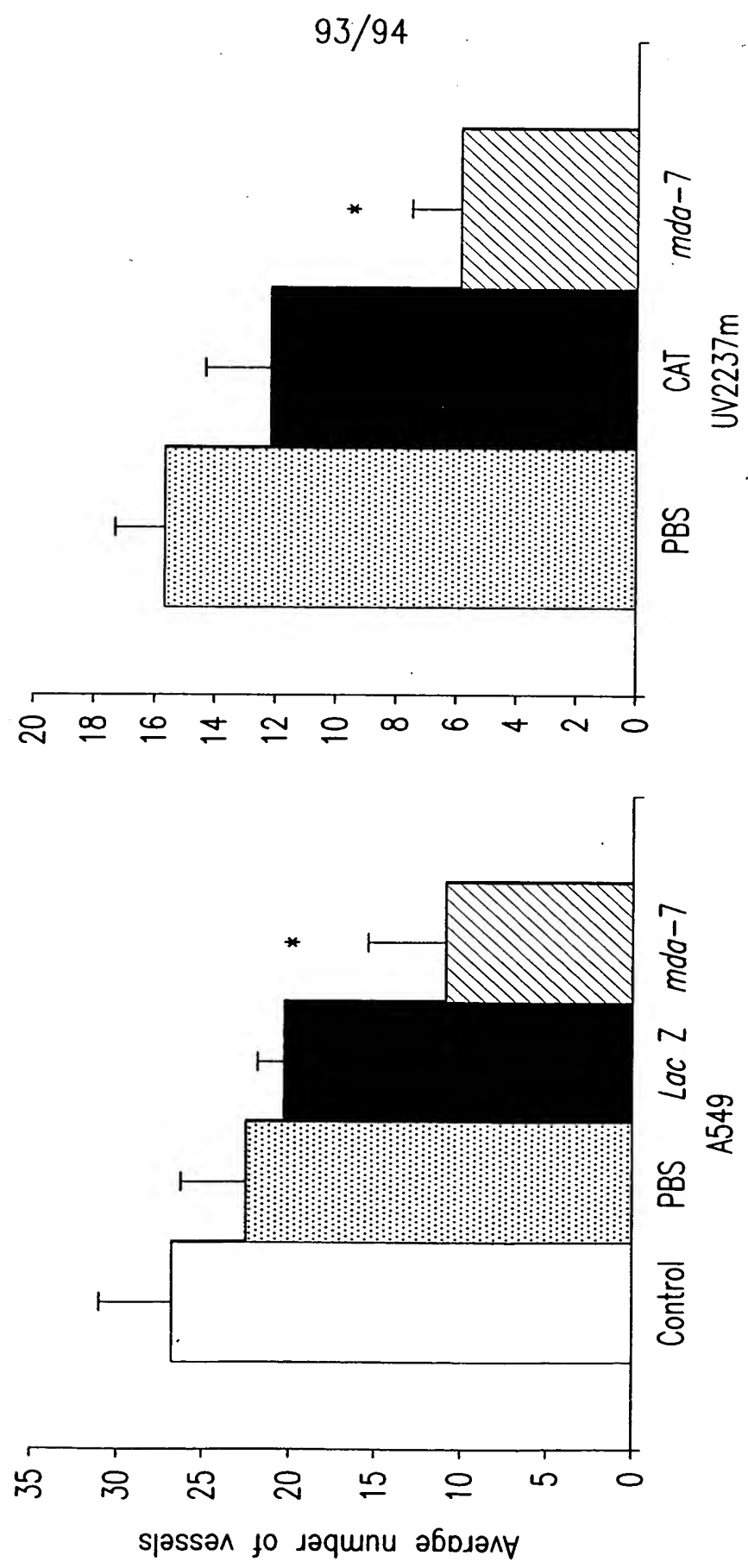


FIG.73

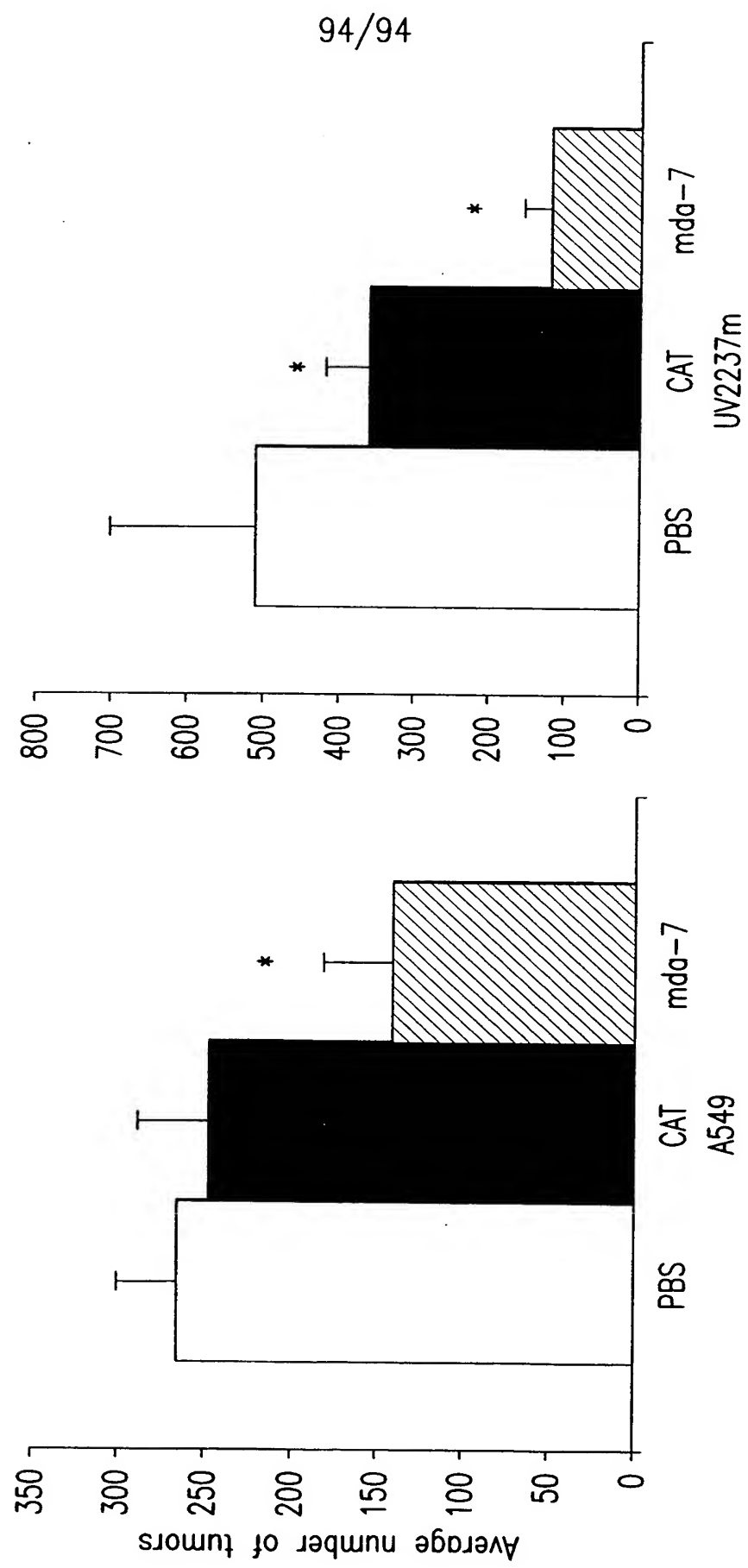


FIG.74

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